

LEVEL ~~III~~

A094742

(14)

YW

# Response of the Cardiovascular System to Vibration and Combined Stresses

AD A110494

PROGRESS REPORT  
1980-1981  
Air Force Office of Scientific Research

~~AFOSR-80-0039~~

Principal Investigator: Charles F. Knapp, Ph.D.

Co-Investigators: Joyce M. Evans, M.S.

David C. Randall, Ph.D.

DTIC  
ELECTED  
FEB 4 1982  
H

DTIC FILE COPY

Wenner-Gren Research Laboratory  
University of Kentucky  
Lexington, Kentucky 40506

November, 1981

82 03 02 089

Approved for public release;  
distribution unlimited.

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

REPORT DOCUMENTATION PAGE		READ INSTRUCTIONS BEFORE COMPLETING FORM
1. REPORT NUMBER <b>AFOSR-TR. 82-0019</b>	2. GOVT ACCESSION NO. <b>AD-A110494</b>	3. RECIPIENT'S CATALOG NUMBER
4. TITLE (and Subtitle) <b>Response of the Cardiovascular System to Vibration and Combined Stresses</b>		5. TYPE OF REPORT & PERIOD COVERED <b>Progress Report 10/1/80 - 9/30/81</b>
		6. PERFORMING ORG. REPORT NUMBER
7. AUTHOR(s) <b>C. F. Knapp, Ph.D., J. M. Evans, M. S. and D. C. Randall, Ph.D.</b>		8. CONTRACT OR GRANT NUMBER(s) <b>AFOSR 80-0039</b>
9. PERFORMING ORGANIZATION NAME AND ADDRESS <b>Wenner-Gren Research Laboratory University of Kentucky, Rose Street Lexington, KY 40506</b>		10. PROGRAM ELEMENT, PROJECT, TASK AREA & WORK UNIT NUMBERS <b>61102F 2312/A1</b>
AFOSR BUILDING 410, BOLLING AFB, D.C., 20332		12. REPORT DATE <b>November, 1981</b>
		13. NUMBER OF PAGES <b>60</b>
		15. SECURITY CLASS. (of this report) <b>Unclassified</b>
14. MONITORING AGENCY NAME & ADDRESS (if different from Controlling Office)		15a. DECLASSIFICATION/DOWNGRADING SCHEDULE
16. DISTRIBUTION STATEMENT (of this Report)  <b>Approved for public release; distribution unlimited.</b>		
17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)		
18. SUPPLEMENTARY NOTES		
19. KEY WORDS (Continue on reverse side if necessary and identify by block number) <b>sinusoidal acceleration, cardiovascular regulation, chronic instrumentation cardiac denervation, total autonomic blockade, ganglionic blockade, left ventricular volume, arginine vasopressin, plasma volume, renin, epinephrine, norepinephrine, cardiac output, stroke volume, heart rate, peripheral resistance</b>		
20. ABSTRACT (Continue on reverse side if necessary and identify by block number) <b>The research effort of the past year was divided into three phases, each of which adds to our systematic investigation of the frequency response character- istics of cardiovascular regulation. The frequency response characteristics are obtained from the analysis of pressure, flow and heart volume responses to low frequency, sinusoidal acceleration (0.001 to 0.25 Hz <math>\pm</math> 2g<sub>z</sub>). Each phase is summarized on the following page.</b>		

DD FORM 1 JAN 73 1473

EDITION OF 1 NOV 65 IS OBSOLETE

UNCLASSIFIED  
SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

- 1) Heart volume measurements in unblocked and autonomically blocked, normal and cardiac denervated dogs undergoing low frequency +2g sinusoidal acceleration; Studies from previous years had indicated an inability of unblocked cardiac denervated, as opposed to normal, dogs to maintain stroke volume during the +g portion of the acceleration stress. To investigate the inadequacy of stroke volume maintenance in these chronically instrumented animals, three cardiac dimensions have been added to provide heart volume information, in addition to our standard instrumentation for measuring aortic flow, left and right ventricular and aortic arch pressures, cardiac output, peripheral resistance, stroke volume, max dp/dt and heart rate. Crystals have been implanted in 44 dogs (21 normal and 23 cardiac denervated). Comparisons of calculated mean volumes with autopsy casts indicated 8 normal dogs had calculated volumes of  $30.6 \pm 5.2\text{cc}$  and cast volumes of  $29.7 \pm 2.6\text{cc}$  while 8 cardiac denervated dogs had calculated volumes of  $26.2 \pm 4.7\text{cc}$  and cast volumes of  $32.8 \pm 2.6\text{cc}$ . Blockades of autonomic effector activity elevated end diastolic volume (n.s.) from  $39.4 \pm 3.6\text{cc}$  to  $47.4 \pm 4.9\text{cc}$  and end systolic volume (sig.) from  $18.5 \pm 2.1\text{cc}$  to  $28.4 \pm 3.6\text{cc}$  in 7 normal dogs. In 4 cardiac denervated dogs end diastolic volumes increased (n.s.) from  $48.8 \pm 8.9$  to  $54.7 \pm 11.6\text{cc}$  with blockade and end systolic volumes increased (sig.) from  $25.9 \pm 7.3$  to  $38.5 \pm 11.6\text{cc}$ . There was a significant decrease (24%) in mean volume of the heart during +g in unblocked normal and cardiac denervated dogs at frequencies below 0.032 Hz with a return toward control volume at higher frequencies. This frequency dependent change in volume was of autonomic origin since it disappeared after autonomic blockade; in cardiac denervated dogs the dependence could not be attributed to neural activity. In normal, unblocked dogs, volumes were significantly greater during -2g than during +2g; a difference which was not evident in unblocked cardiac denervated dogs.
- 2) Comparison of autonomic effector blockade with ganglionic blockade; In an effort to determine the best hydraulic "nonreflexive" cardiovascular system for study, 6 animals underwent our centrifuge protocol after blockade of  $\alpha$ ,  $\beta$  and muscarinic effector sites and again after addition of ganglionic blockade to the effector blockade. Results of this study indicated that 1) The ganglionic blockade lowered heart rate and increased stroke volume when added to the autonomic blockade but had no effect on cardiac output, peripheral resistance or aortic pressure. 2) The post-acceleration increase in peripheral resistance but not aortic pressure was greater with ganglionic as opposed to effector blockade. 3) In general, the ganglionic blockade is thought to more closely approximate the hydraulic "nonreflexive" aspect of the circulatory system than does autonomic effector blockade.
- 3) The effect of cardiac denervation on cardiovascular function before and after autonomic effector blockade; After several years of using cardiac denervated animals in our acceleration studies we have summarized the effects of cardiac denervation per se. In unblocked "reflexive" dogs cardiac denervation results in decreased plasma renin activity (sig.), norepinephrine (sig.), epinephrine (n.s.) and plasma volume (n.s.). In this state there was no difference in aortic pressure, right ventricular diastolic pressure, cardiac output, heart rate, stroke volume, peripheral resistance, plasma osmolality or vasopressin. After autonomic effector blockade, cardiac denervated animals were found to have lower "intrinsic" heart rates (sig.) and higher stroke volumes (sig.) and maintained lower levels of plasma volume, renin, norepinephrine and epinephrine but these differences were no longer significant.

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

## TABLE OF CONTENTS

Form DD 1473 (Abstract)

### Introduction

I. CARDIAC DIMENSION CHANGES DURING DYNAMIC ACCELERATION LOADINGS IN NORMAL AND CARDIAC DENERVATED DOGS.....	3
Methods.....	4
Surgical Procedures.....	4
Crystal Fabrication and Testing.....	4
Analog Equipment Modifications.....	7
Digital Data Acquisition and Analysis.....	10
Analog Volume Computer.....	13
Experimental Protocol.....	16
Procedure for Autonomic Blockade.....	17
Test Sequence.....	18
Preliminary Results and Conclusions.....	18
Changes in Left Ventricular Volume in Control States....	18
Sinusoidal Acceleration Response.....	22
II. COMPARISON OF TOTAL AUTONOMIC EFFECTOR BLOCKADE WITH GANGLIONIC BLOCKADE.....	34
Results.....	34
Conclusions.....	41
III. THE EFFECT OF CARDIAC DENERVATION ON CARDIOVASCULAR FUNCTION BEFORE AND AFTER AUTONOMIC EFFECTOR BLOCKADE.....	43
Methods.....	43
Surgical.....	43
a) Cardiac denervation.....	43
b) Implant procedures.....	43
c) Postoperative care.....	44
Experimental Protocol and Blood Sampling Techniques.....	44
a) Plasma renin activity.....	45
b) Plasma arginine vasopressin activity and osmolality.....	45
c) Plasma norepinephrine and epinephrine activity.....	45
d) Plasma volume and hematocrit.....	45



## INTRODUCTION

The experiments conducted during the past year were performed to expand our knowledge of the frequency response characteristic of cardiovascular regulation. In these studies, low frequency, sinusoidal acceleration ( $0.001$  to  $0.25 \text{ Hz}$ ,  $\pm 2g_z$ ) was used as the non-invasive input disturbance to the cardiovascular system. In the past we have described the acceleration-induced oscillatory behavior of the cardiovascular system without neural feedback control and the less oscillatory behavior of the intact system with full regulatory control of the heart and peripheral vasculature. The animal preparation without neural feedback control was originally produced by pharmacologic blockade of  $\alpha$ ,  $\beta$  and muscarinic receptors. To this blockade we now have added hexamethonium to block nicotinic receptors as well. The rationale for this phase of the experiment was based on the large outpouring of catecholamines in response to autonomic blockade which might be exerting a tonic, unblockable stimulation of autonomic effectors due to large concentrations in the synaptic cleft. A comparison of acceleration-induced responses of the same animal with and without the addition of hexamethonium to our standard pharmacologic blockade ( $\alpha$ ,  $\beta$ , and muscarinic receptors) is presented in Section II.

The unblocked or regulated responses to acceleration disturbances have also been examined further during the present year. A major part of this endeavor has been to dissect the regulatory capabilities of the cardiovascular system by examining the responses of animals who have been cardiac denervated. This intervention produced an animal preparation with the cardiac output component of regulation deprived of neural feedback control, but with the peripheral vasculature totally innervated. Sections I and III deal specifically with this endeavor, in particular Section III deals with the hemodynamic

and neurohormonal responses to cardiac denervation per se while Section I examines the response of denervated dogs as compared to normal dogs during acceleration. In this section the emphasis is on the differences and similarities in the heart volume response of these dogs to autonomic blockade as well as to acceleration.

The experiments reported herein were conducted according to the principles described in "Guide for the Care and Use of Laboratory Animals," DHEW Publication No. (NIH) 78-23.

# I. CARDIAC DIMENSION CHANGES DURING DYNAMIC ACCELERATION LOADINGS IN NORMAL AND CARDIAC DENERVATED DOGS

Results from our previous studies have indicated that an intact cardiac regulatory mechanism was essential for maintaining the integrity of the cardiovascular system during low frequency, sinusoidal acceleration stress. Specifically, the inability of cardiac denervated (as opposed to fully innervated) dogs to maintain blood pressure during the  $+2 g_z$  portion of the acceleration stress was found to be principally due to the inability of these animals to maintain stroke volume during this time. Mechanisms proposed to explain the inadequacy of stroke volume maintenance included: a smaller end diastolic volume in cardiac denervated dogs implying disconnection of a) an afferent (sensory) mechanism which adjusted venous return or b) an efferent mechanism responsible for adjustment of diastolic (relaxed) heart size, or both. Alternatively, a large end systolic volume at the same or increased end diastolic volume would imply disconnection of an efferent mechanism for adjustment of end systolic volume (the standard sympathetic inotropic pathway). Any or all of these mechanisms could combine to result in a decrease in stroke volume, and for that reason we implemented a procedure to provide direct information about volume changes of the left ventricle in response to acceleration stress. The technique chosen was that of Rankin et al (1) in which pairs of ultrasonic dimension crystals were implanted to record major and minor epicardial dimensions and wall thickness. From these data, the volume was calculated assuming a prolate ellipsoidal geometry. What follows is a description of our progress to date in: surgical implant procedures; crystal fabrication and testing; analog equipment modifications to allow for simultaneous measurement of electromagnetic flow, pressure and dimension; modification of instrumentation for centrifuge compatibility; digital data acquisition techniques and results from the experiments conducted so far.



## Methods

### Surgical procedures:

Forty four mongrel dogs (23 normal and 21 cardiac denervated) were instrumented for further study. Each animal was anesthetized with sodium thiopental 20 mg/kg, intubated, positive pressure respirated, and an L4 thorotomy performed under sterile conditions. In those animals who were denervated, the procedure detailed in Sect. III, Surgical Methods was done at this time. In these animals and in the normally innervated animals, a pair of 5 mm diameter ultrasonic dimension transducers was then positioned to record the maximum anterior to posterior epicardial left ventricular chamber diameter. Another 5 mm pair was positioned to give epicardial major axis diameter (one crystal was placed in the groove between the left atrium and the pulmonary artery and the other was located near the apical dimple). Wall thickness of the left ventricle was measured by a 1 mm diameter crystal tunneled to the endocardial surface of the anterior wall and a 3 mm diameter crystal positioned directly over it. Each pair of crystals was aligned at surgery to give the best received signal possible, and further monitoring was done following placement of the remaining instrumentation (left ventricular pressure gauge, aortic flow probe, right atrial cannula) to ensure continued signal quality. Placement of the other transducers is detailed in Section III. The chest wall was closed, the leads placed in the subcutaneous dacron pouch and the animal was allowed at least three weeks of postoperative recovery. In two animals, the leads were exteriorized at the time of surgery in order to monitor changes in heart shape and size postoperatively.

### Crystal fabrication and testing:

The transducers used for making dimension measurements were made from type LTZ 2, Lead Titanate Zirconate (Transducer Products), 5 MHz, "thickness mode" plates of piezoelectric ceramic to which was added a defocusing lens on

the front surface. Construction of the transducers was begun by breaking the 5 MHz crystal material into 4 mm and 1 mm squares. The corners were then rounded on a lathe or with fingernail clippers and emery board. In order for the wires to be soldered properly, the crystal was cleaned with acetone and the area to be soldered scraped with a scalpel. The wires to be soldered to the crystal were tinned so that only a quick touch of the soldering iron was required to affix the wire to the material.

The crystals were cleaned further with denatured alcohol, detergent (Ecosil), and rinsed in deionized water before and after epoxy was applied. This process helped to eliminate electrically conductive contaminants. An epoxy resin (Stylcast, W. R. Grace Co.), which was used to make the lens for the transducer, was mixed with its hardener and placed in a vacuum chamber until completely degassed. The resin mixture was then placed in a mold to produce 4 mm diameter lenses. A lens was glued to the crystal and then a 6 mm length of 1/8" diameter shrink tubing was slid over the transducer and heated just enough to shrink the tubing. The transducer and 5 mm of lead were then immersed in epoxy and placed in a vacuum chamber. To accommodate suturing to the heart surface, the transducer was sandwiched with silastic between a backing of closed cell foam and a front patch of Dacron. To promote optimal signal transmission a 5 mm hole was made for the lens in the center of the Dacron patch.

The 1 mm crystals were slightly different in that thin, stainless steel wire was used and was threaded through a length of tubing to assist in placement through the heart wall. Also, both sides of the crystal material were lensed, because transducer orientation could not be controlled during surgical placement on the endocardial surface of the left ventricle.

Testing of the finished transducer was accomplished by affixing the transducers to movable posts and measuring signal strength at various rotation angles

and also by measuring signal fallout as the transducers were moved to the edges of the transmitted signal until voltage output of the receiving transducer decreased to .1 volt. Work is also continuing to improve our understanding of transducer performance. A move has been made away from the laser Schlieren system used last year because it appears that the representation of the continuous wave excitation of these crystals required for the Schlieren system may not be the same as that from the pulse-transit excitation used to generate the cardiac dimension during our studies. Constructive and destructive interference caused by the multi-frequency pulse-transit signal may make the actual beam pattern differ from the continuous wave pattern used for the Schlieren system. Therefore, the signal falloff method mentioned above is the one currently in use.

A calibration test has been performed on the Schuessler Model 401 Ultrasonic Dimension System which revealed an error in the calibration of the recorded data. In that test were two identical ultrasonic transducers of the type used for chronic implantation (flat-type, 4 mm diameter 5 MHz LTZ with convex lens 1 mm thick). Chart paper was calibrated using the Schuessler Model 401 built-in calibration and LED display. The major axis channel #1 was calibrated from 40 mm - 80 mm full scale since this is typically the distance range in which it is used. Likewise, the minor axis channel #2 was calibrated from 25 mm - 65 mm full scale. Wall thickness channel #4 was calibrated from 5 mm - 25 mm full scale. Next the transducers were mounted in an adjustable jig that would allow the transducers to be moved to an exact distance apart as measured with a vernier caliper. It was assumed that when a transducer is chronically implanted its lens is pushed 0.5 mm into the heart surface. Therefore, it was necessary to allow for this distortion when measuring the distance between the transducers. A signal was recorded using the Schuessler

Model 401 at 5 mm distance intervals as measured with the vernier caliper. The recorded signal of the known distance was compared to the built-in calibration of the Schuessler Model 401. These measurements indicated that the Schuessler Model 401 outputs a distance greater than that measured by the vernier caliper for each channel tested. When major, minor and wall thickness dimensions are corrected as per the vernier caliper reading the resulting calculated volume (Figure I-1) was found to be only slightly less than the uncorrected value. Note that in the volume range of 20cc-30cc the present values are only 2-3cc higher than the corrected values, however, as the volumes increased the error also increases.

Therefore, it appears that in our range of volumes, the reduction in volume resulting from the reduced major and minor axis readings is approximately offset by the increase in volume resulting for the reduction in wall thickness. This produced a smaller error (range 0-8%) than had been initially expected.

In the future, hemispherical ultrasonic transducers may be used such as the technique of van Trigt, et. al., (2) which consists of a hemispheric crystal with a quarter-wavelength matched face plate. The result is a beam pattern and alignment that is greatly improved over the flat type crystal. Hemispheric transducers have a beam width of  $+90^\circ$  as compared to about  $\pm 20^\circ$  with the flat type transducer. Since the beam angle is wider for the hemispheric transducer, surgical placement would be more accurate.

#### Analog equipment modifications:

As expected, simultaneous use of the dimension crystals with the electromagnetic flow transducers produced signals which interfered with each other. Conversations with other researchers and with the manufacturers of our electromagnetic square-wave flow meter (Zepeda Instruments) and our dimension meter (Schuessler and Associates) resulted in two suggestions:

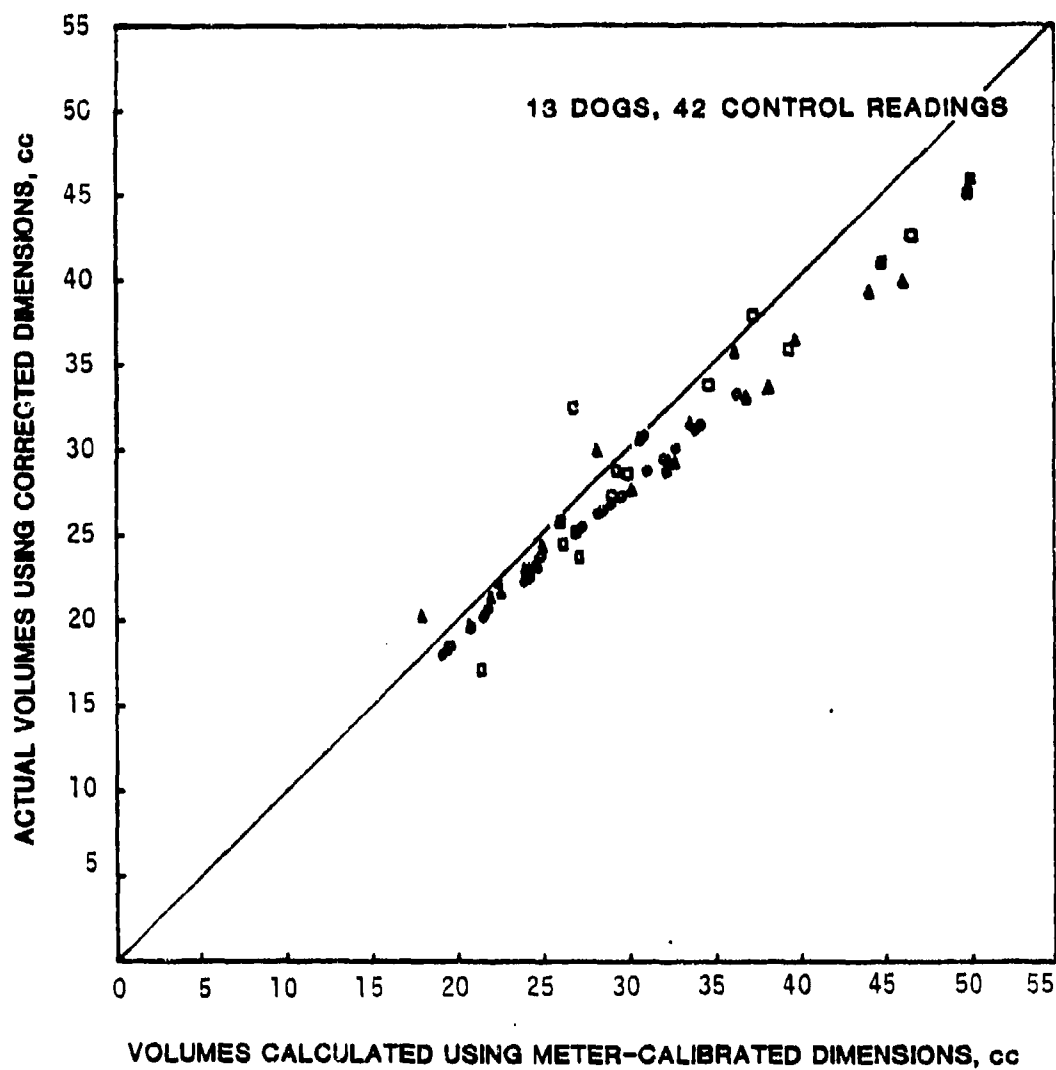


FIGURE I-1. Comparison of calculated volumes using the corrected dimension readings (Y Axis) as opposed to volumes calculated from direct, uncorrected, readings. (X Axis)

1) an outline of possible steps to take to electronically synchronize the two pieces of equipment or 2) manually sample from first one meter and then from the other and assume that the same experimental conditions hold from one time to the next. Due to the nature of our dynamic experimental environment, the second suggestion was rejected and several attempts to synchronize our equipment were made.

The initial suggestions from the manufacturers resulted in a digital circuit which synchronized the electromagnetic drive oscillator and flow gate signal to certain divisions of the dimension clock frequency. This circuit consisted of a divider circuit, a pulse-width and signal shaping circuit and a low-impedance driving circuit. This device took a signal from the dimension meter at a point in the dimension circuit that had already been divided and after further division by the new circuit, this signal was used to override and capture the electromagnetic oscillator. This synchronizing circuit worked nicely for synchronizing at certain frequencies, but for those frequencies which are necessary for operation of the flowmeter oscillator (a limited operating band) only a portion of the interference could be eliminated. Additionally, the flowmeter gate was not adjustable, further limiting our timing ability.

Given the features of the Zepeda flowmeter in design (square-wave) and accessibility (sealed assemblies) we decided to move to another flowmeter of different design and circuitry. The Biotronix 610 pulsed logic flowmeter which we have used for several years was designed to operate at frequencies near that required for synchronization. Also, schematics were available for its circuit components, and those components were not sealed. After studying the design of this flowmeter, it appeared that its digital circuitry would be more compatible with the digital component circuitry of the dimension meter. Finally, the design of its flow gate was such as to allow for sequential excitation

of all four dimension transmitter pulses and the associated crystal ring and therefore make possible the removal of ultrasonic interference from the electromagnetic flow signal (Figure I-2).

With these considerations in mind, a circuit was bread-boarded which used the dimension meter clock pulse (divided down) as the flow oscillator, rather than capturing the flow oscillator as was necessary with the Zepeda meter. This circuit plus an adjustment of the flowmeter resulted in simultaneous dimension and electromagnetic flow readings of excellent quality (see results section). This circuit is basically an analog inverter made from a general purpose operational amplifier with a frequency response high enough to pass an undistorted clock pulse, Figure I-3. These synchronized meters were then modified for mounting on the centrifuge platform and have become a part of our standard centrifuge instrumentation package.

Ultrasonic interference with the pressure signals was not of the same magnitude and therefore no further corrective action was taken.

#### Digital data acquisition and analysis:

Calculations of heart volume were made on an off-line basis using a PDP 11/34 computer. All analog signals including the three heart dimension measurements were recorded on an 14 channel Ampex tape recorder during each experiment. The recorded physiological data were sampled at two millisecond intervals and smoothed with a digital filter. The geometry of the left ventricle was represented as a three-dimensional, prolate ellipsoidal shell. The following assumptions about the ellipsoidal shell were made: 1) the measured minor and major axis diameters were assumed to represent the external diameter of the shell, 2) the measured anterior wall thickness was used as the dynamic shell thickness at the minor axis circumference, 3) the shell thickness at the base and apex was assumed to be 55% of the equatorial value

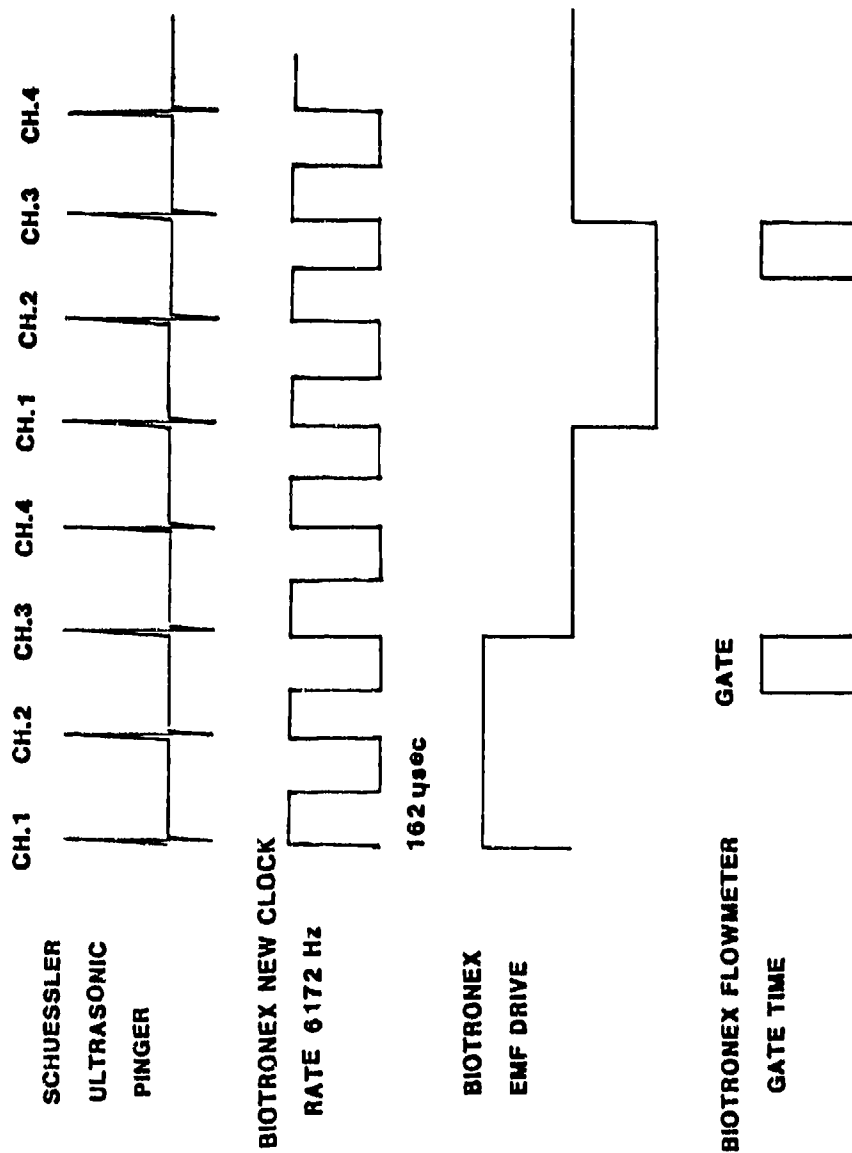


FIGURE I-2

Timing diagram for synchronization of ultrasonic dimension transmitter pulses to electromagnetic flow driver and gate



# FLOWMETER SYNC DRIVER

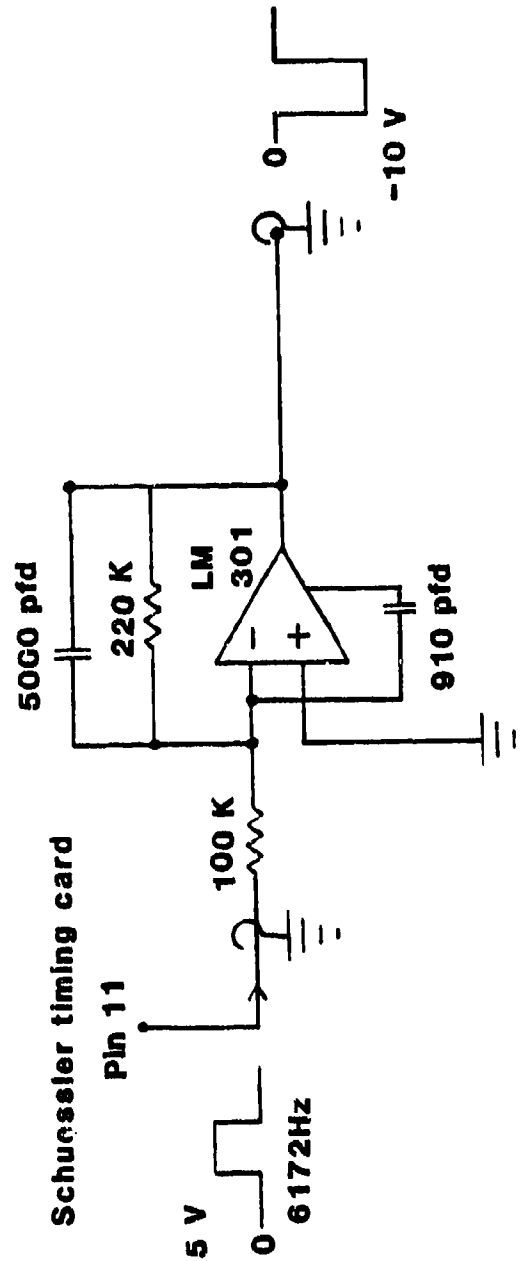


FIGURE I-3

Schematic of flow-meter synchronization driving circuit

(based on measurements on postmortem hearts). The dynamic internal volume of the shell was computed using the formula for the prolate ellipsoid:

$$V = \frac{\pi}{6} (b-2h)^2 (a-1.1h)$$

where  $b$  is the external minor axis diameter,  $h$  is the equatorial wall thickness, and  $a$  is the external major axis diameter. Stroke volume was computed as the change in internal shell volume during ejection. Stroke volume was also calculated by integration of the aortic flow curve assuming zero flow at end-diastole. The stroke volume measured with the flowmeter can also be compared to the data calculated from the dimension measurements.

Differentiation of the calculated volume curve can then be used to determine inflow and outflow from the left ventricle and again outflow can be compared to the electromagnetic flow trace. For left ventricular pressure-volume plots, left ventricular pressure was filtered in the same manner as that for volume and plotted against the corresponding value of left ventricular pressure.

#### Analog Volume Computer

An analog circuit was also constructed to compute left ventricular volume from the above equation for a prolate ellipsoid:  $vol = \pi/6(b-2h)^2 (a-1.1h)$ . Figure I-4 is a block diagram containing several components which will be discussed separately. Starting with the three dimension output signals from the Schuesslermeter, each signal was passed through an offset biasing circuit which is shown in detail in Figure I-5. The component biased each input signal to voltage levels which represent actual dimensions measured. With each dimension now in the same voltage range, the wall thickness signal for the major axis ( $1.1h$ ) was derived from the minor axis wall thickness ( $2h$ ) by an inverter/component shown in detail in Figure I-6. At this point, the four signals  $2h$ ,  $b$ ,  $1.1h$ , were sent to a subtractor circuit (left hand side, Figure I-7)

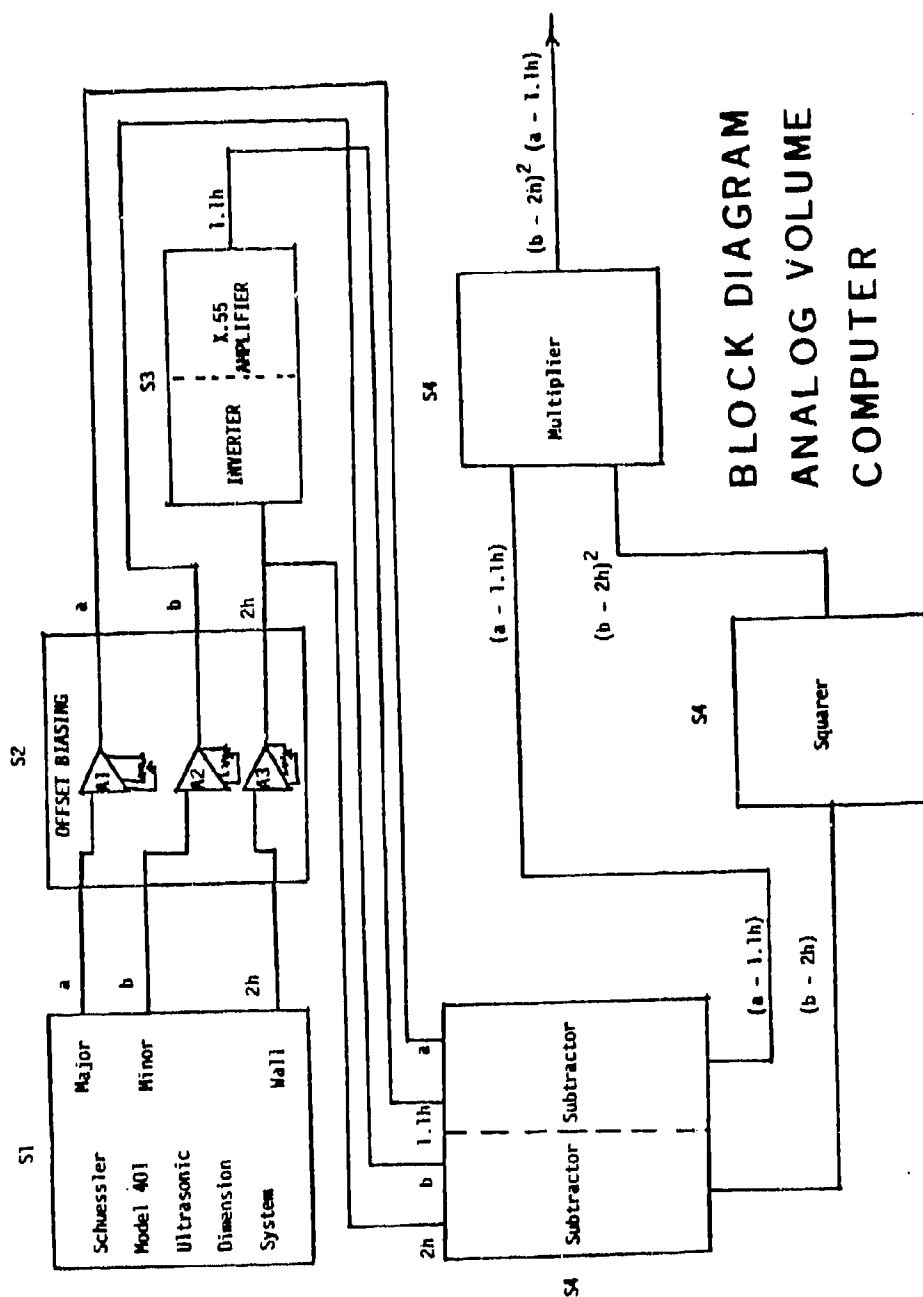


FIGURE I-4 Block diagram of the analog volume computer used in the present study. Individual components of this circuit are shown in detail in FIGURES 5,6, and 7.

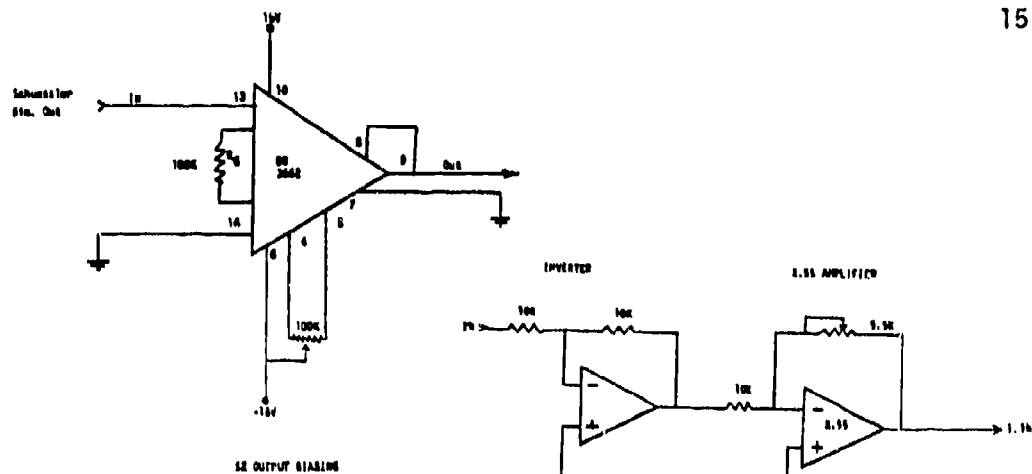


FIGURE I-5 Offset biasing circuit component of the analog volume computer.

FIGURE I-6 Constant multiplier component of the analog volume computer.

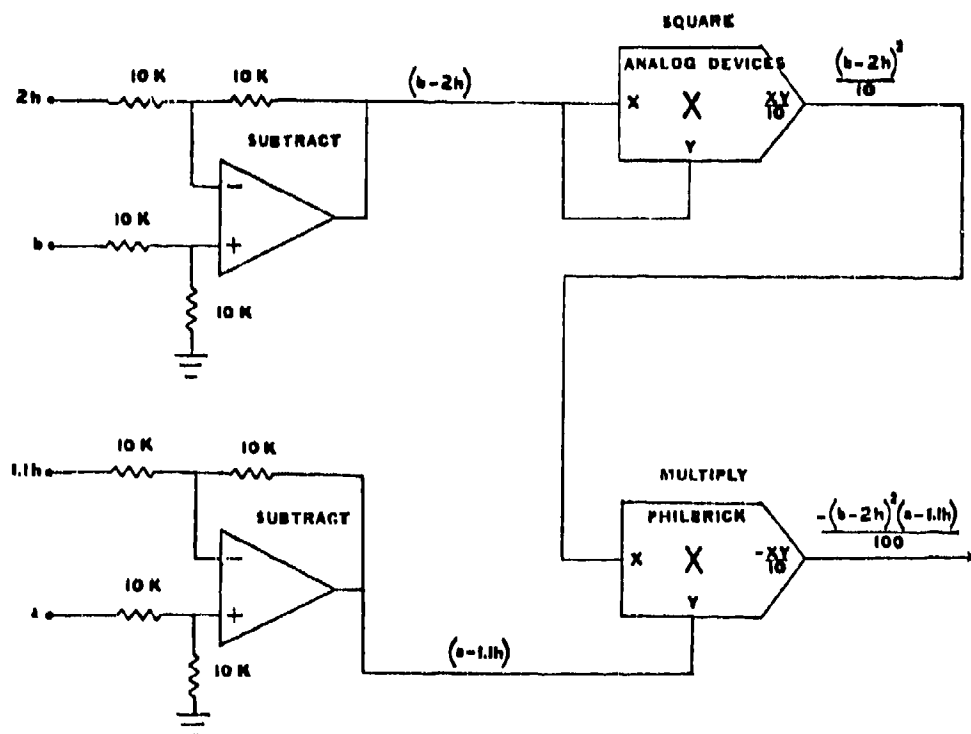


FIGURE I-7 Final subtractor, multiplier and squaring components of the analog volume computer.

resulting in (b-2h) and (a-1.1h). The (b-2h) signal was then squared (upper right hand corner, Figure I-7). The constant  $\pi/6$  was disregarded and final scaling is done by the chart recorder. This analog circuitry has been used on tape replays to provide a continuous volume output for data analysis and could be used on-line, but accuracy would be questionable since final readings of each dimension, particularly wall thickness, cannot be determined until autopsy.

#### Experimental Protocol

On the day of the experiment, the animal was tranquilized with an intramuscular injection of Innovar Vet at 0.075 cc/kg. Piezoelectric manometer-tipped catheters (Millar PC 350, 5 French) were placed, under local anesthetic, in the right and left ventricles via small branches of a main femoral vein and artery, respectively. The arterial Millar gauge was used to calibrate the implanted Konigsberg gauge and then retracted into the aorta, just outside the aortic valve, to measure arterial pressure. The animal was maintained in a lightly tranquilized state for the duration of the experiment with serial injections of Innovar (0.5 cc/hr) administered through the right atrial cannula.

The measured physiological variables included aortic pressure and flow, left and right ventricular pressure and heart rate. In addition, measurements of left ventricular, major and minor axis dimensions and wall thickness have been added as described above. On-line, digitally calculated variables included beat-by-beat stroke volume, cardiac output, peripheral vascular resistance, maximum dp/dt, and the pressure difference from the aorta to the right atrium. Left ventricular volume is either calculated digitally off-line or on-line by an analog computer circuit.

### Procedures for Autonomic Effector and Ganglionic Blockades

In order to delineate the neural and nonneural components of the measured cardiovascular responses to acceleration, a pharmacologically-induced total autonomic blockade was used to inhibit adrenergic and cholinergic activity at the effector site, thus removing normal reflex barostatic action. The increase, however, in circulating catecholamines by two orders of magnitude, the tripling of both plasma renin and vasopressin resulting from the establishment of this blockade and the enhanced pressor response to acceleration in the presence of this blockade led us to a comparison of the autonomic effector blockade with a ganglionic blockade; these differences are detailed in Section II.

The total autonomic blockade for the proposed study consisted of the alpha adrenergic blocker phenoxybenzamine (Dibenzylamine) at 20 to 30 mg/kg administered over an hour, beta blockade with propranolol (Inderal) at 1 to 2 mg/kg over approximately ten minutes, and cholinergic blockade with atropine (Atropine Sulphate) at 0.1 to 0.2 mg/kg over approximately five minutes. The efficacy of the blockade was tested and verified by a comparison of systemic responses to specific agonists given prior to blockade, following blockade and then again at the conclusion of the blocked acceleration sequence. These consisted of a 40 ug/kg bolus of phenylephrine (Neosynephrine) to test the alpha blockade and a 0.5 ug/kg bolus of isoproterenol (Isuprel) to test the beta blockade. If heart rate showed evidence of reflexive parasympathetic activity (i.e., a decrease following phenylephrine) the atropine dosage was supplemented.

The ganglionic blockade was established after the acceleration tests were performed on the animals with the total autonomic blockade. Administration of hexamethonium at 10 mg/kg over a 10 minute period was used, and the ganglionic blockade was antagonized before and after acceleration with the alpha and beta agonists as above.

### Test Sequence:

The test sequence and experimental conditions were the same as in the previous years to allow for correlation of the experimental results. The sequence consists of  $\pm 2 g_z$  sinusoidal acceleration from 0.004 to 0.3 Hz starting at the low frequency and moving to the next higher frequency at 3 to 4 minute intervals without stopping the centrifuge. Previous years' studies indicated that this protocol allowed steady state conditions to develop more rapidly than did a protocol which stopped the centrifuge between frequencies. The sinusoidal series of tests was followed by a step input series consisting of 3 minutes each from  $+ 2 g_z$  to  $- 2 g_y$  to  $- 2 g_z$  to  $+ 2 g_y$  to  $+ 2 g_z$  to  $- 2 g_z$  to  $+ 2 g_z$  and back to  $- 2 g_y$ . The first 4 inputs were produced by  $90^\circ$  rotation of the platform while the centrifuge is producing 2 g radial acceleration. The last 2 inputs were produced by a  $180^\circ$  rotation each.

### Preliminary Results and Conclusions

#### Left Ventricular Volumes in Control States

Calculations of left ventricular volumes for the various control states resulting from our experimental protocol are shown in Table I for all animals studied, and in Figure I-9 for those animals who completed all phases of the study. In Table I, 21 normal dogs were instrumented with an average heart volume of  $23.9 \pm 3.1$  cc at surgery while 23 cardiac denervated dogs were instrumented with a post-denervation volume of  $29.5 \pm 3.0$  cc at surgery. Three weeks later, at the time of the centrifuge study, heart volume for a group of 10 normal animals averaged  $28.6 \pm 3.6$  cc and for a group of 10 denervated animals averaged  $31.3 \pm 4.1$  cc. Subsequent beta adrenergic blockade increased heart size of the normal animals to  $36.8 \pm 3.7$  cc and of the denervated animals to  $36.3 \pm 6.0$  cc. Subsequent muscarinic blockade had little effect on heart volumes in either group. Addition of alpha adrenergic blockade to complete

	NORMAL				CARDIAC DENERVATED			
	N = 21				N = 23			
	MAJOR mm	MINOR mm	WALL mm	VOLUME cc	MAJOR mm	MINOR mm	WALL mm	VOLUME cc
$\bar{x}$	69	53.2	12.6	23.9	67.1	53	11.4	29.5
S.E.M.	1.5	1.0	.7	3.1	.9	1.3	.6	3.9
	N = 10 UNBLOCKED				N = 10			
$\bar{x}$	70.8	52.9	11.5	28.6	68.5	55.7	11.8	31.3
S.E.M.	2.7	1.5	.6	3.6	1.1	2.2	.6	4.1
	N = 9 BETA BLOCKADE				N = 6			
$\bar{x}$	73.1	55.5	10.9	36.8	68.9	56.8	11.3	36.3
S.E.M.	2.5	1.1	.6	3.7	1.3	2.9	.5	6
	N = 8 BETA & MUSCARINIC BLOCKADE				N = 6			
$\bar{x}$	72.9	55.1	11.3	33.9	69.8	58.1	11.1	37.9
S.E.M.	2.9	.9	.7	3.4	1.3	2.6	.6	6.1
	N = 8 TOTAL AUTONOMIC BLOCKADE				N = 6			
$\bar{x}$	72.8	55.5	11.4	34.4	70.1	58.9	11.3	41.9
S.E.M.	2.8	1.05	.7	3.4	1.1	2.8	.7	9.9
	N = 3 GANGLIONIC BLOCKADE				N = 4			
$\bar{x}$	71.5	54.2	11.4	31	66.8	55.5	11.8	29.1
S.E.M.	1.3	1.4	.8	5.7	.8	2.5	.8	3.1
	N = 10 POSTMORTEM (IMMEDIATE)				N = 10			
$\bar{x}$	71.9	55.9	10.9	39.9	70.5	56.5	11	37.5
S.E.M.	2.4	1.6	.9	5.9	1.6	2.4	.6	4.7
	N = 8 POSTMORTEM (12 HOURS)				N = 8			
$\bar{x}$	68	54.4	11.3	30.6	66.8	54.1	12.1	26.2
S.E.M.	1.8	1.5	.7	5.2	2.0	2.3	.64	4.7
	N = 7 CAST (12 HOURS POSTMORTEM)				N = 10			
$\bar{x}$				29.7				33.7
S.E.M.				2.6				2.6

TABLE I. Comparison of dimension measurements and calculated heart volumes for normal dogs and cardiac denervated dogs at sequential stages of the experimental protocol.



the total blockade resulted in a further increase in heart volume in cardiac denervated dogs and little change in size in normal dogs. Sacrificed heart volumes were the largest of any volumes in the normal dogs and were large, but not the largest, volumes observed in denervated dogs. At autopsy (12 hrs. after sacrifice) heart volumes for both groups had returned to near unblocked control volumes. At this time, a silastic cast of the ventricular chamber was made and the volume of water displaced by the cast was recorded. It was felt that a comparison of the cast volume to the 12 hours postmortem calculated volume would be the most accurate comparison possible, and resulted in a calculated volume of  $30.6 \pm 5.2$  cc as compared to a cast volume of  $29.7 \pm 2.6$  cc for 7 normal dogs and a calculated volume of  $26.2 \pm 4.7$  cc and a cast volume of  $33.7 \pm 2.6$  for 10 cardiac denervated dogs.

A plot of changes in mean heart volume for the group of animals (8 normal, 6 cardiac denervated) who completed all phases of the study are shown in Figure I-8. The results for this group are quite similar to the larger group (Table 1). Since all subjects in these two groups received all treatments, changes with treatments can be discussed with less chance of error. From this plot, both normal and cardiac denervated animals are seen to increase mean heart size from the time of surgery to experiment. This was expected due to the open-chest, anesthetized state of the animal at the time of surgery. On the day of the experiment, beta adrenergic blockade increased heart volume (significantly in normal animals), addition of muscarinic blockade did not further change heart volume but total blockade increased volume further. Sacrificing of the animal with pentobarbitol resulted in the largest volumes seen in both normal and cardiac denervated dogs. Again both calculated and cast, postmortem volumes were near unblocked control values. Three normal dogs and four cardiac denervated dogs (data not shown) were treated with the

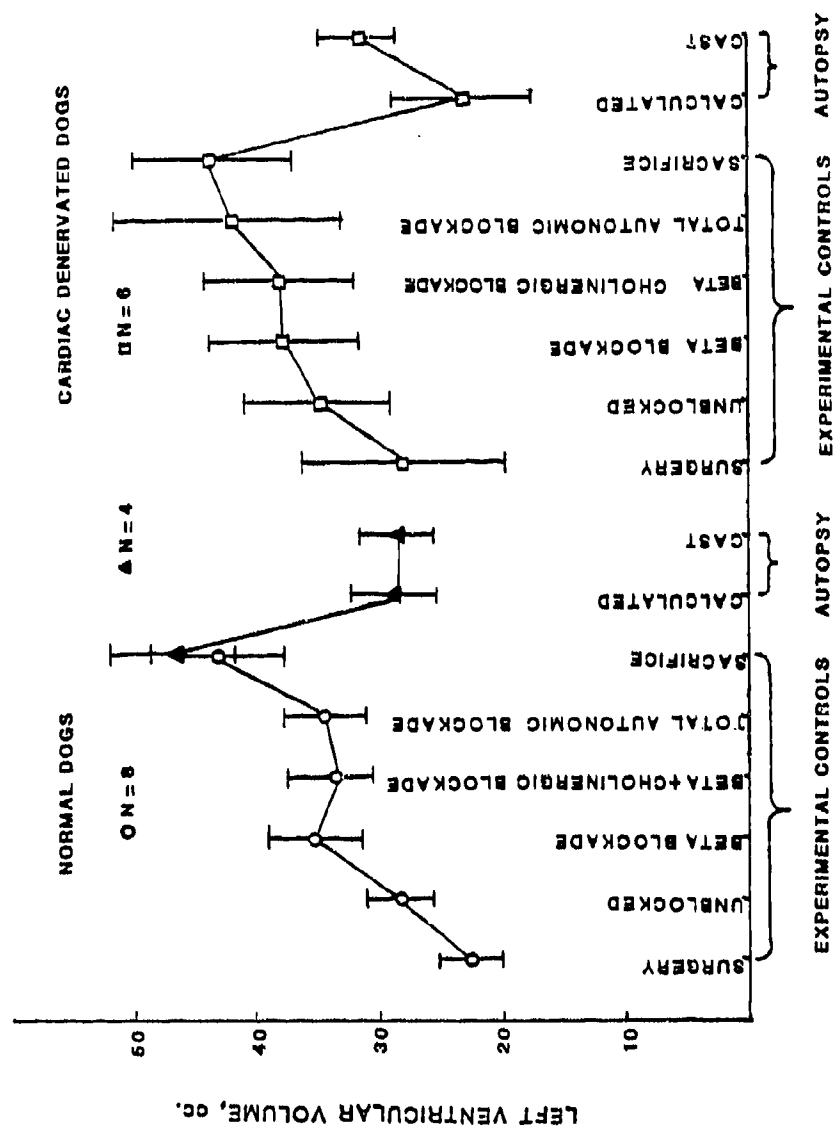


Figure I-8 Comparison of calculated heart volumes for a group of 8 normal dogs with a group of 6 cardiac denervated dogs who completed all stages of the experimental protocol.

ganglionic blocker hexamethonium after the total autonomic blockade; in the normal animals, heart volumes were unchanged from the totally blocked volumes. And in the cardiac denervated dogs, volumes were down [ $23.8 \pm 2.9$  cc to  $29.1 \pm 3.1$  cc] but not significantly. Conclusions from this segment of the study are:

1. Cardiac denervation does not significantly change the mean volume of the left ventricle at time of surgery or three weeks later at the time of study.
2. Beta adrenergic blockade increases mean left ventricular volume by 25% in normal dogs and by 6% in cardiac denervated dogs.
3. Addition of muscarinic and alpha adrenergic blockade to the beta blockade does not further increase heart size in normal dogs but does increase volume another 15% in cardiac denervated dogs.
4. The 50% drop in stroke volume resulting from establishment of total autonomic blockade (1980 progress report) is due to loss of effector activity and is not a result of decreased filling of the left ventricle since blockade elevated heart volume by 20% in normal dogs and by 40% in cardiac denervated dogs.

#### Sinusoidal Acceleration Response

##### a. Normalization

After postmortem comparison of cast heart volumes to calculated heart volumes, the best retrograde adjustment possible was made for each dimension reading at each state of the experiment. To date, dimension measurements have been made on 10 normal and 10 cardiac denervated dogs undergoing our centrifuge protocol (See Figure I-9). Two slightly different analyses of heart volume changes during acceleration have been tried, both on an off-line basis. The first, a continuous calculation of volume, using either analog or digital techniques, gave a record of volume throughout the total

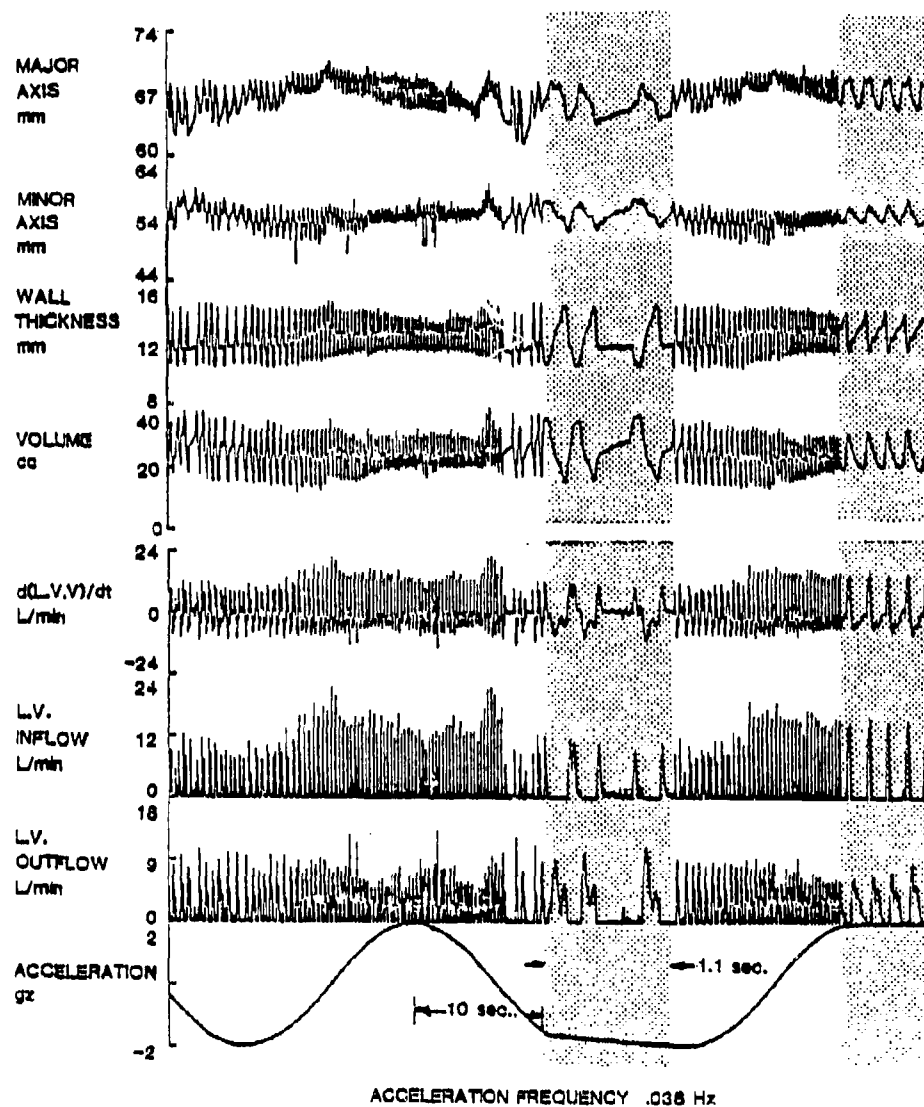


FIGURE I-9 Cardiac dimension and calculated dimension parameters for a single animal during  $+2G_z$  acceleration.

experiment. This analysis indicated slight changes in volume in the sacrificed animal that could be interpreted as actual volume changes due to acceleration-induced pressure differences across the valves of the heart. An alternative explanation could be the coupling of surgical implant adhesions with acceleration to produce changes in the shape of the ventricle which would also be present in the beating heart. Since there was no indication of flow from the aortic flow probe, the shape change was felt to be more likely and therefore the changes seen in the animal after sacrifice were used to normalize the changes seen in the live animal.

The left ventricular volume normalizing procedure normalizes with respect to left ventricular volume during the sacrificed animal acceleration run. By using the least squares method, the equation for the difference between the mean volume and the volume at the point of interest, ( $+g_y$ ,  $-g_y$ ,  $+g_z$ , or  $-g_z$ ) versus the log of the acceleration frequency, during the sacrificed animal run were determined. Using this equation the difference at each frequency of the live animal run is estimated. The estimated difference is subtracted from the live animal data, which is then stored as the normalized volume data.

For illustration, volume data for two frequencies before and after a similar normalization is shown in Figure I-10. This normalization procedure consisted of several steps, the first of which was analyzing left ventricular chamber's wall thickness ( $h$ ), and both its major ( $a$ ) and minor ( $b$ ) axes for frequency dependent variations during acceleration of the dead dog. The dead dog data was not found to be frequent dependent. Two cycles of acceleration, wall thickness, and major and minor axes were sampled from the dead dogs data at a 3.1 msec. rate. This data was then used for the normalization of the live dog data. The live dog was sampled at 6.2 msec. for both a high and low acceleration frequency. The following formula was used to calculate

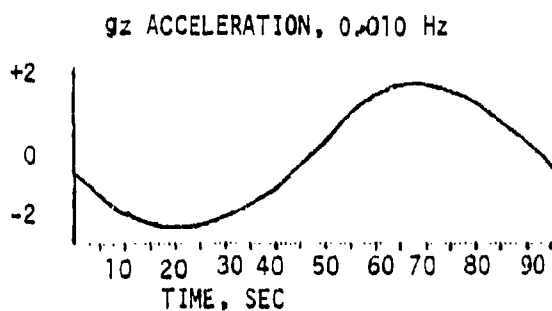
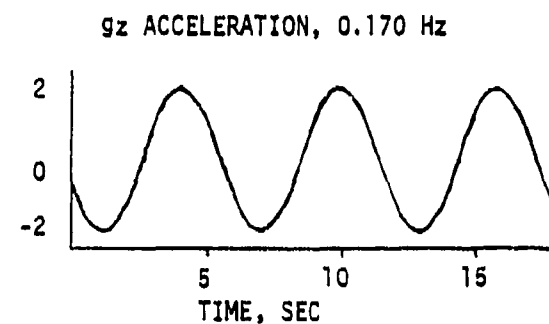
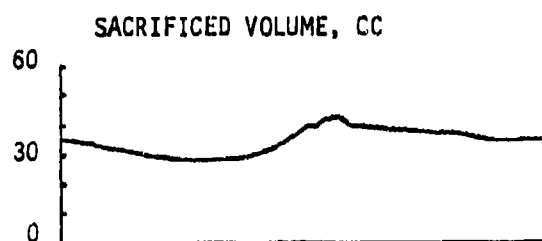
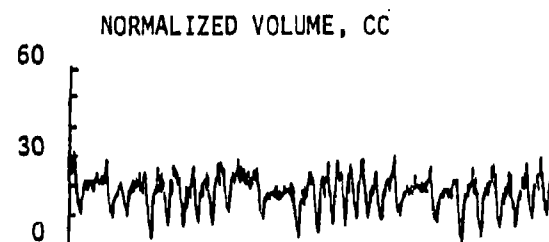
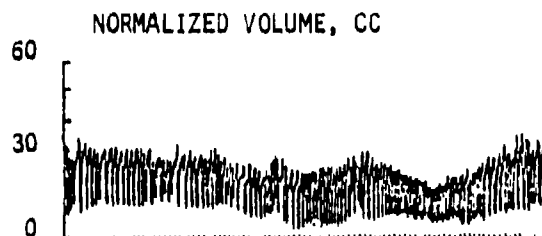
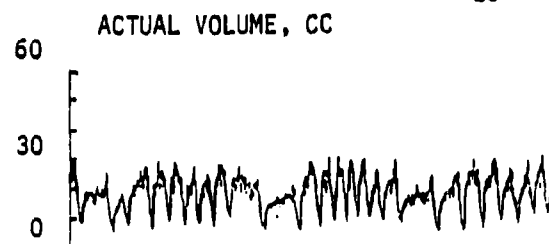
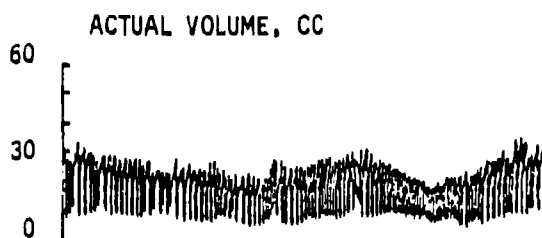


FIGURE I-10. Two examples of off line, digital normalization of actual left ventricular volumes by changes seen in the sacrificed dog undergoing the same acceleration exposures. See text for details of normalization.

left ventricular chamber volumes for the dead dog and the two live dog accelerations:  $v = \pi/6(b-2h)(a-1.1h)$ . The dead dog volume used to normalize the low frequency live dog volume was placed in phase with the live dog data. Linear interpolation was used to expand the dead dog data to the same number of points per cycle as the live dog data. Dead dog and live dog data were of the same frequency for the high frequency test. The normalization data for the high frequency test was created by using every other point from the dead dog volume data and repeating the data three times. The following formula was used to normalize the two live dog accelerations:  $v_n = v_l - (v_d - \bar{v}_d)(\bar{v}_l / \bar{v}_d)$ , where  $v_n$  = normalized volume,  $v_d$  = dead dog volume, and  $v_l$  = live dog volume.

On continuous playback analysis this posed a considerable off-line signal processing problem if done accurately with quite a large chance for error if done by hand. An alternative analysis has proved to be accurate and much less time consuming. At four points in the acceleration cycle (+2gz, -2gz, +2gy, and -2gy) data is taken from each of the three dimension channels and a volume is calculated. The same procedure is done in the animal postmortem and changes from the mean at each of these points are then either added or subtracted from the live dog data at the same points depending on whether or not the values were above or below the mean. It should be emphasized that these changes are normally quite small when compared to the total acceleration-induced changes seen in the live animal.

#### b. End Diastolic and Systolic Volumes

In 7 normal and 4 cardiac denervated animals the heart volume data was analyzed before and after acceleration in both the unblocked and totally blocked state for end diastolic and end systolic volumes, Figure I-11. For each pair of bars, the open bars are pre-acceleration means  $\pm$  SEM and

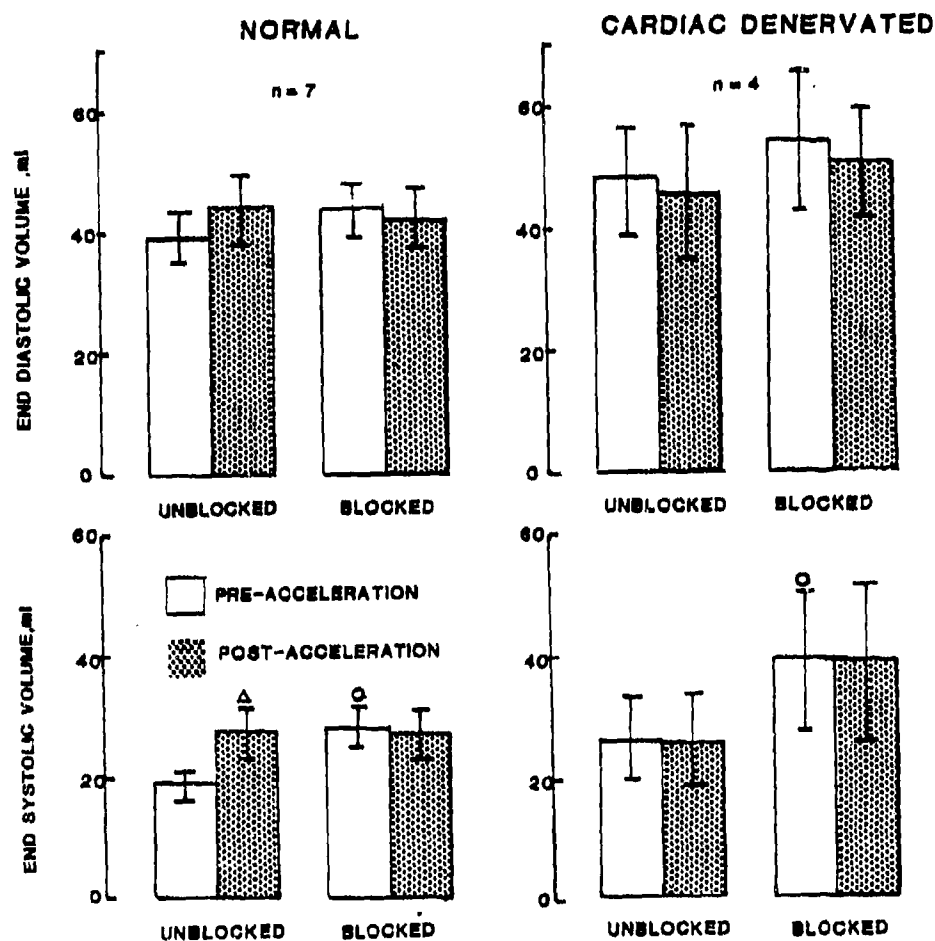


FIGURE I-11. Mean  $\pm$  SEM pre-acceleration (open bars) and post-acceleration (stippled bars) end diastolic volumes (above) and end systolic volumes (below) for 7 normal and 4 cardiac denervated dogs in the unblocked state and following blockade of  $\alpha$ ,  $\beta$ , and muscarinic effector activity. Significant changes with acceleration are indicated by triangles while significant changes with blockade are indicated by circles.



the stippled bars are post-acceleration mean  $\pm$  SEM for the same animals in either the unblocked or autonomically blocked state.

End diastolic volumes are shown in the top half of the figure and end systolic volumes are in the bottom half. Normal animals in the unblocked state had an average end diastolic volume of  $39.4 \pm 3.6$  ml before acceleration and  $44.0 \pm 5.6$  ml after acceleration. After blockade, end diastolic volume increased to  $47.4 \pm 4.9$  ml for these animals and returned toward control values ( $42.2 \pm 4.4$  ml) at the end of the acceleration. Cardiac denervated dogs in the unblocked, pre-acceleration state had slightly larger end diastolic volumes,  $48.8 \pm 8.9$  with no real change ( $46.2 \pm 10.8$ ) as a result of acceleration. Autonomic blockade of these cardiac denervated dogs increased heart size to  $54.7 \pm 11.6$  ml and again acceleration returned these blocked end diastolic volumes toward control ( $51.1 \pm 8.9$  ml). Overall there was no statistically significant change in end diastolic volume with either denervation, autonomic blockade or acceleration.

End systolic volumes for the same animals did however change significantly with both acceleration and autonomic blockade. Normal, unblocked animals increased (triangles,  $p > .05$ ) end systolic volume from  $18.5 \pm 2.1$  ml to  $27.0 \pm 4.2$  ml as a result of acceleration. Autonomic blockade of these animals also increased (circles,  $p > .05$ ) end systolic volumes to  $28.3 \pm 3.6$  ml and acceleration of these blocked animals produced no further significant change ( $27.2 \pm 3.6$  ml). Cardiac denervated animals did not change ( $25.9 \pm 7.3$  ml) systolic volume with acceleration but did significantly increase systolic volumes with blockade ( $38.5 \pm 11.6$  ml) with no further change resulting from acceleration.

The results of this segment of the study indicate that 30 minutes of sinusoidal acceleration to unblocked, normally innervated dogs results

in increased end diastolic (non significant) and end systolic (significant) volumes of the left ventricle. From earlier studies (1980-1981) progress report) we have observed a beta adrenergically mediated increase in heart rate and vasodilation in response to the same stress. It appears that this activation of the beta adrenergic system is selective for heart rate and vasodilation since there was no increase in stroke volume noted in the present study.

The increase in end systolic volume in both normal and cardiac denervated animals in response to autonomic effector blockade implicates these pathways as being critically important for maintenance of stroke volume in normally innervated and cardiac denervated dogs. In normally innervated dogs this could be neurally and/or hormonally mediated while in cardiac denervated dogs with neural pathways ablated, efferent activity is confined to hormonal activation of the beta adrenergic activity.

In unblocked cardiac denervated dogs, the lack of increase in either end diastolic or end systolic volume in response to acceleration further implicates a neural pathway for the response seen in neurally intact dogs. Finally, while not statistically significant, cardiac denervated dogs operated around larger end diastolic volumes in all cases than did normal dogs and in all cases but one (post-acceleration, unblocked) they also had larger end systolic volumes.

c.+2g<sub>z</sub> and -2g<sub>z</sub> unblocked responses: As detailed in the normalization section above, average +2g<sub>z</sub> volumes (□) and -2g<sub>z</sub> volumes (Δ) for six normal (Figure 1-12a) and four cardiac denervated (Figure I-12b) dogs are shown as a function of acceleration frequency. Starting from approximately the same average control values ( $28.4 \pm 2.8$  and  $26.7 \pm 5.0$  ml), both normal and cardiac denervated dogs' mean heart volumes significantly decreased by 24% during +2g<sub>z</sub> at the lowest frequencies. In normal dogs, the +2g<sub>z</sub> heart volumes remained significantly lower than control at the 4 lowest frequencies, then climbed back toward control values as accel-

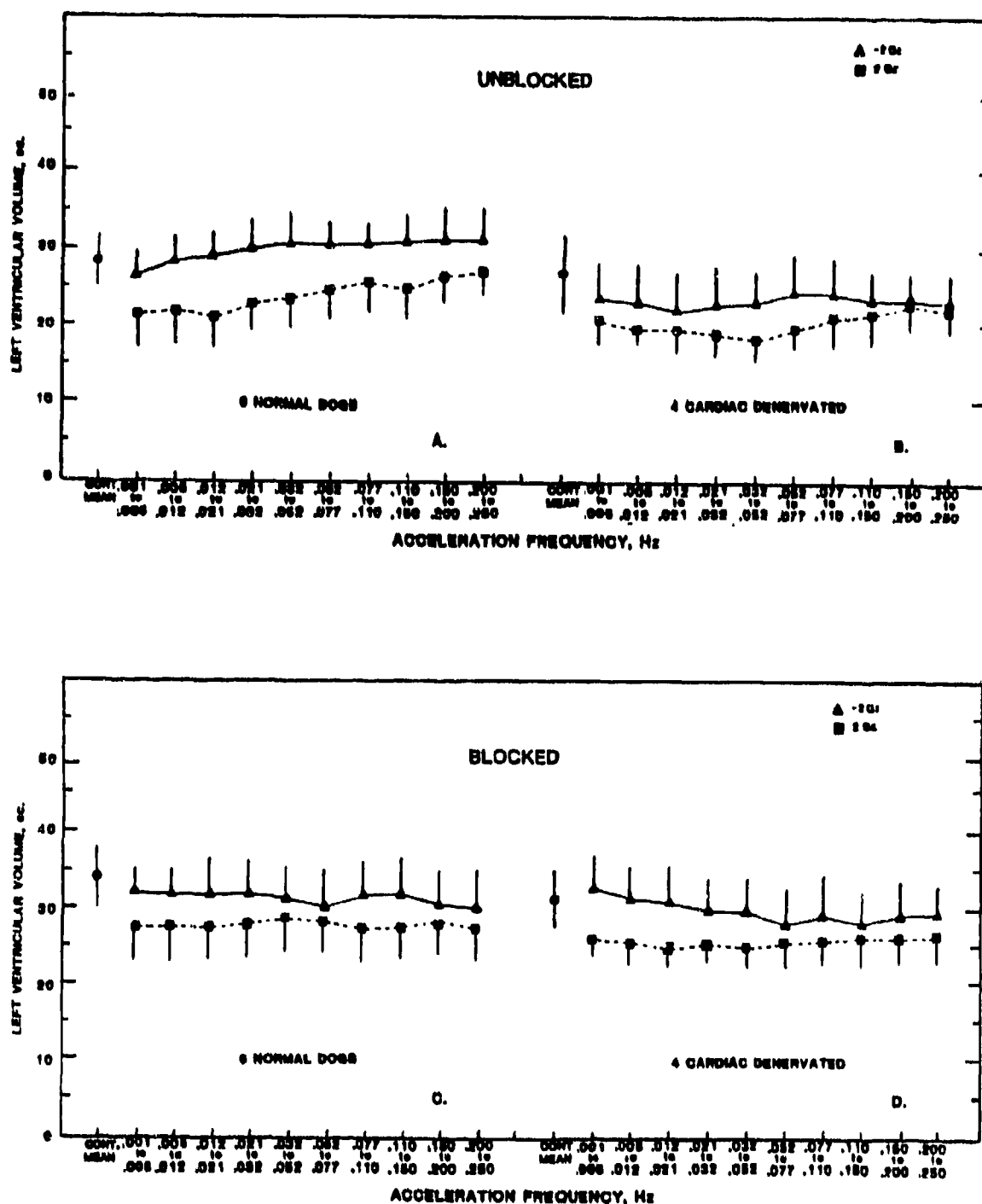


FIGURE I-12A,B,C,D. Comparison of +2gz and -2gz ventricular volumes in response to sinusoidal acceleration in (A) normal dogs in the unblocked state; (B) cardiac denervated dogs in the unblocked state; (C) normal dogs after autonomic effector blockade; and (D) cardiac denervated dogs after autonomic effector blockade.

ation frequency increased, reaching control values at the highest frequencies. The increase in  $+g_z$  volume across the frequency range was significant in that values up to .02 Hz were lower than values above 0.1 Hz. Cardiac denervated dogs maintained the 24% decrease in heart volume during  $+2g_z$  up to 0.052 Hz and then begin to return toward control but without ever reaching that volume even at the highest frequencies. The  $-2g_z$  response for both normal and cardiac denervated dogs were different, cardiac denervated dogs did not indicate any frequency dependent changes, while  $-g_z$  values climbed significantly between the first and last frequencies in normal dogs. There was another difference between the two groups: the normal dogs'  $-2g_z$  heart volumes remained elevated at or above control across the frequency range, while in cardiac denervated dogs the values remained below control across the frequency range. Finally the differences between the  $+g_z$  and  $-g_z$  were significant at each frequency except the last one in normal dogs and at four of the five lowest frequencies in cardiac denervated dogs.

d.  $+2g_z$  and  $-2g_z$  totally blocked responses: Figures I-12c and I-12d show the  $+$  and  $-2g_z$  results from the same 6 normal dogs (I-12c) and 4 cardiac denervated dogs (I-12d) undergoing the same acceleration test, following total autonomic blockade. In this condition, acceleration lowered heart volumes significantly below control in both groups of animals with no discernable frequency dependent changes across the 0.005 to 0.25 Hz frequency range. In the normal dogs,  $-g_z$  values were also below control values but not significantly and there was no frequency effect. In the cardiac denervated dogs the  $-g_z$  values at the lowest frequency were significantly elevated above those at 0.052 Hz and above; this "frequency effect" is probably a time effect since it was the tendency of all animals, particularly cardiac denervated ones, to return toward unblocked control heart volumes with time after blockade which

produced in itself a larger heart volume than did the acceleration stress. In both groups of animals in the blocked state,  $-g_z$  volumes were significantly greater than  $+g_z$  volumes at frequencies below 0.032 Hz.

Conclusions from this segment of the study concerning heart volumes are:

1. Autonomic effector blockade significantly increased heart volume in all animals in control and during  $+g_z$  and  $-g_z$  acceleration.
2. The unblocked, "reflexive" response of both normal and cardiac denervated dogs to  $+2g_z$  stress shows a frequency dependence across the range studied (0.005 to 0.25 Hz). In both groups of animals heart volumes significantly decreased below control levels at low frequencies and came back toward control with increasing frequency.
3. The  $+2g_z$  frequency dependence is a result of autonomic effector activity since this frequency dependence disappears following autonomic effector blockade. In cardiac denervated dogs this response cannot be attributed to neural activity.
4. The response of normal dogs to  $-2g_z$  across the frequency range 0.005 to 0.25 Hz indicated a frequency dependence in which values increased with increasing frequency in unblocked dogs. This response was not present in blocked normal dogs or cardiac denervated dogs in either unblocked or autonomically blocked states.
5. The  $-2g_z$  increase in volume above control in normal animals appeared to be due to autonomic effector activity that was of neural origin since it was not present in unblocked cardiac denervated dogs or in normal dogs following autonomic blockade. This implies a neurally mediated relaxation of the left ventricle in response to increase venous return.

## REFERENCES

1. Rankin, J. S. et al. The three-dimensional dynamic geometry of the left ventricle in the conscious dog. *Circ. Res.* 39 (3): 304-313. 1976.
2. Van Trigt, Peter, III, Bruce J. Bauer, Craig O. Olsen, J. Scott Rankin, and Andrew S. Wechsler. An improved transducer for measurement of cardiac dimensions with sonomicrometry. *Am. J. Physiol.* 240 (Heart Circ. Physiol. 9): H664-H668, 1981.

## II. COMPARISON OF TOTAL AUTONOMIC EFFECTOR BLOCKADE WITH GANGLIONIC BLOCKADE

In an attempt to delineate the components of the mean pressor response associated with  $\pm 2g_z$  sinusoidal acceleration after blockade of autonomic effector activity, we added blockade of nicotinic receptors to our previously established autonomic blockade. This blockade, using hexamethonium at 10 mg/kg, blocks the principle transmission of neural signals at all autonomic (both sympathetic and parasympathetic) ganglia. The rationale for this phase of the experiment was based on the large outpouring of catecholamines in response to autonomic blockade (specifically the alpha blocker, dibenzylamine) which at the time was felt perhaps to be exerting a tonic, unblockable stimulation of autonomic effectors due to large concentrations in the synaptic cleft. By blocking the ganglionic transmission of neural efferent signals, it was reasoned that release of catecholamines at post ganglionic synaptic terminals would be reduced and that circulating levels of catecholamines would return toward unblocked levels. Preliminary analysis of catecholamine levels has shown this to be the case, and the results will be reported when enough studies have been done.

Our standard sinusoidal acceleration protocol was used with the ganglionically blocked animals and a comparison of the results with those from the same animals with autonomic effectors blocked are presented below. Details of the animal preparation and experimental protocol are given in Section III.

### RESULTS

The first and most consistent observation concerning the animals in the ganglionically blocked state was that oscillations in aortic pressure in these animals were more sinusoidal than those of the same animals after effector blockade. Figure II-1 shows the mean (top panel), maximum and minimum

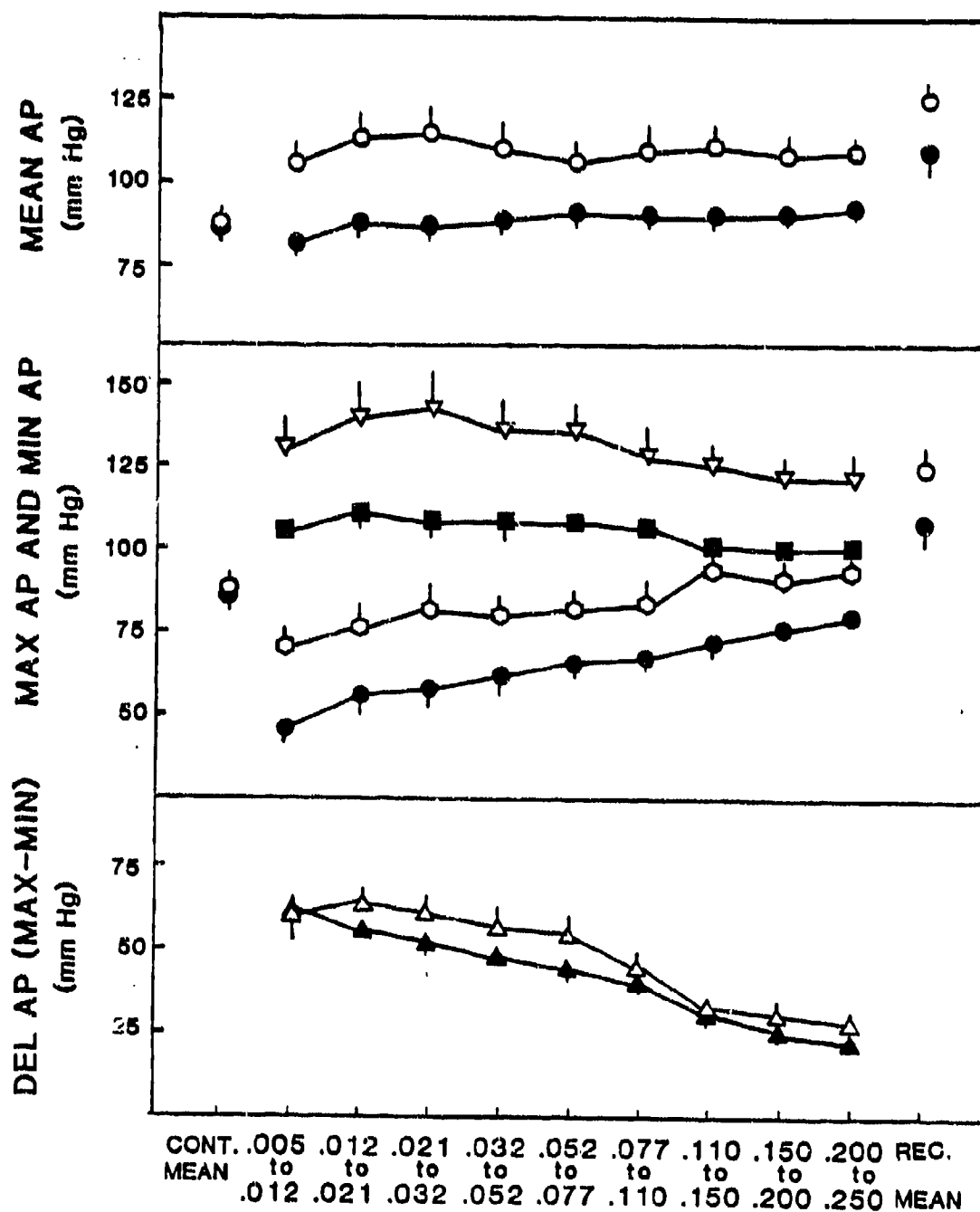


FIGURE II-1. Comparison of the aortic pressure response to acceleration for 6 dogs after autonomic effector blockade (open symbols) and ganglionic blockade (closed symbols).



(middle panel), and delta (maximum-minimum) aortic pressure response to acceleration for a group of 6 animals after autonomic effector blockade (open symbols) and following addition of ganglionic blockade (closed symbols). The mean pressor response in these two states was quite different. First there was not significant pressor response during acceleration of ganglionically blocked dogs, although in the post acceleration recovery phase one developed, while in the effector blocked dogs, a pressor response was evident within the first few minutes of acceleration. Due to oscillation around a lower mean pressure, ganglionically blocked aortic pressures dropped lower in the  $+g_z$  phase and did not go as high in the  $-g_z$  phase when compared to the effector blocked response. Finally, division of the low frequency bin into 2 bins [.003 to .0065] and [.0065 to .012] (not shown) indicated that the magnitude of DEL AP in effector blocked dogs was constant up to about 0.052 Hz before dropping off at higher frequencies while the magnitude of DEL AP in the ganglionically blocked dogs peaked at the lowest frequencies, dropping off with each increase in frequency. Cardiac output responses for the two groups are shown in Figure II-2. Control cardiac outputs for the two groups were quite similar [ $2.6 \pm .2$  L/min] for the autonomic effector blockade compared to  $2.5 \pm .4$  L/min for the ganglionic blockade. In both groups there was a frequency dependent change in mean values of cardiac output starting from a low of  $1.8 \pm .3$  L/min at the lowest frequencies and rising to  $2.1 \pm .1$  L/min for the ganglionic and to  $2.4 \pm .3$  L/min for the autonomic effector group at the highest frequencies. Maximum and minimum responses for the two groups are remarkably similar across the frequency range. What difference there is in the maximum and minimum between the two groups is apparent in the DEL CO response where DEL CO for the effector blocked group did not change with frequency but dropped with increasing frequency for the ganglionically blocked group.

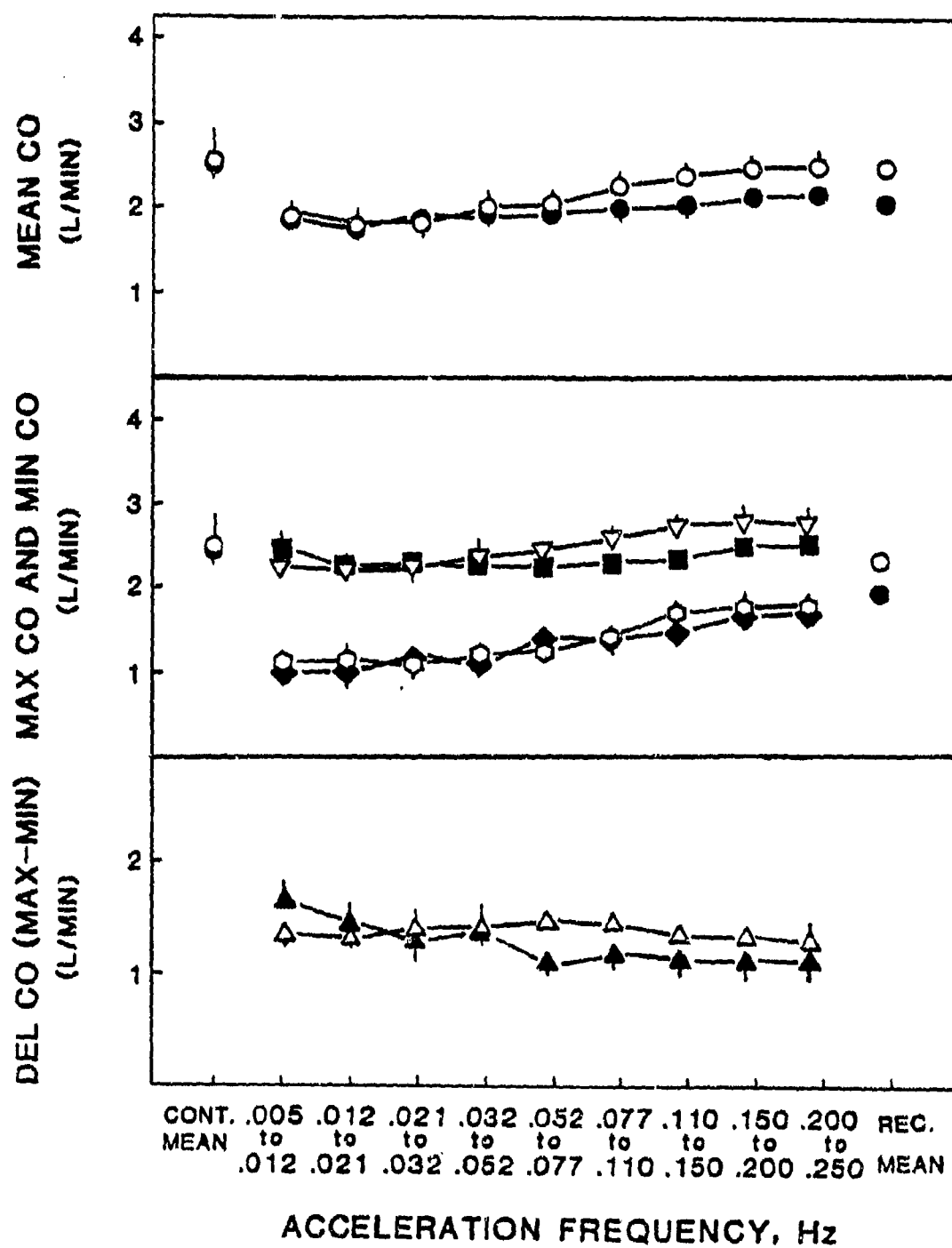


FIGURE II-2. Comparison of the cardiac output response to acceleration for 5 dogs after autonomic effector blockade (open symbols) and ganglionic blockade (closed symbols).

The heart rate response of the two groups was not different. Due to the blockades, there was no change in heart rate with acceleration, even though there was a difference in the control values. Heart rates were lower [ $114 \pm 5$  b/min] for the ganglionic as compared to the effector [ $135 \pm 7$  b/min] blockade. Since there was no change with acceleration, the heart rate data will not be shown.

Comparison of stroke volume responses between the two groups is shown in Figure II-3. Control values of stroke volume were higher in the ganglionic [ $19.5 \pm 1.9$ cc] as compared to the autonomic effector [ $16 \pm 2.5$ ] blockade state. There was a frequency dependent increase in mean stroke volume with the lowest stroke volumes occurring at the lowest frequencies and rising with increasing frequencies back toward, but not reaching, control values. The drop in mean stroke volume in both groups was associated with the  $+2g_z$  portion of the acceleration cycle with no increase in stroke volume over control values during the  $-2g_z$  portion of the cycle. There was a frequency dependent change in DEL SV (decreasing DEL SV with increasing frequency) in the ganglionically blocked dogs but not in the dogs with effector blockade. Peripheral resistance, the pressure drop across the systemic circulation divided by cardiac output ( $\Delta P/Q$ ), is shown in Figure II-4. For both the total autonomic effector and the ganglionic blockades, acceleration produced an increase in peripheral resistance that was maintained across the frequency range and was elevated in post-acceleration recovery. The pre- to post-acceleration change in resistance with effector blockade [34 to 51] mmHg/(L/min)] was smaller than that with ganglionic blockade [30 to 58 mmHg/(L/min)]. But, the change in the mean value of resistance during acceleration was larger in the totally blocked dogs than in the ganglionically blocked dogs due primarily to lower minimum values of

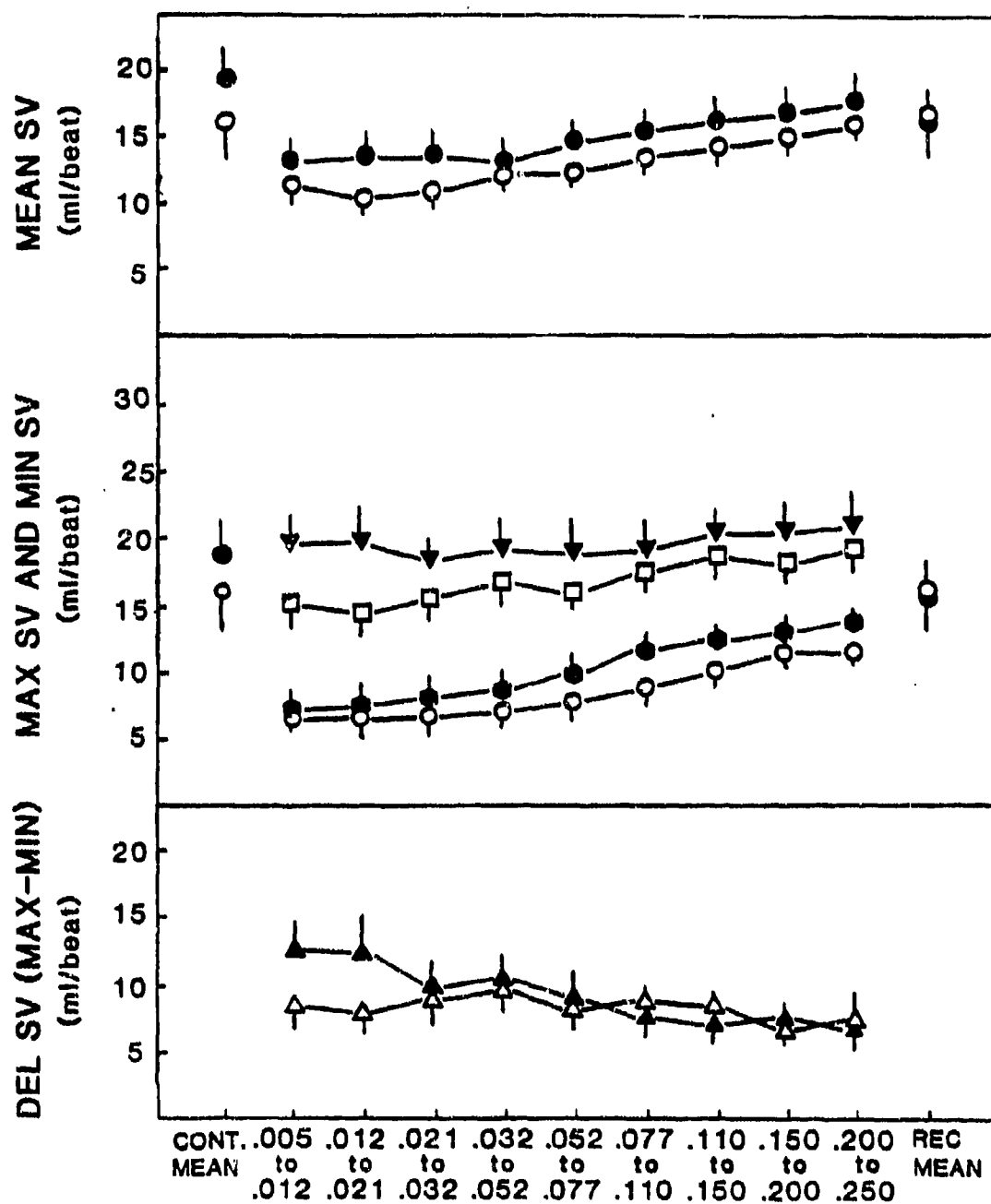


FIGURE II-3. Comparison of the stroke volume response to acceleration for 5 dogs after autonomic effector blockade (open symbols) and ganglionic blockade (closed symbols).

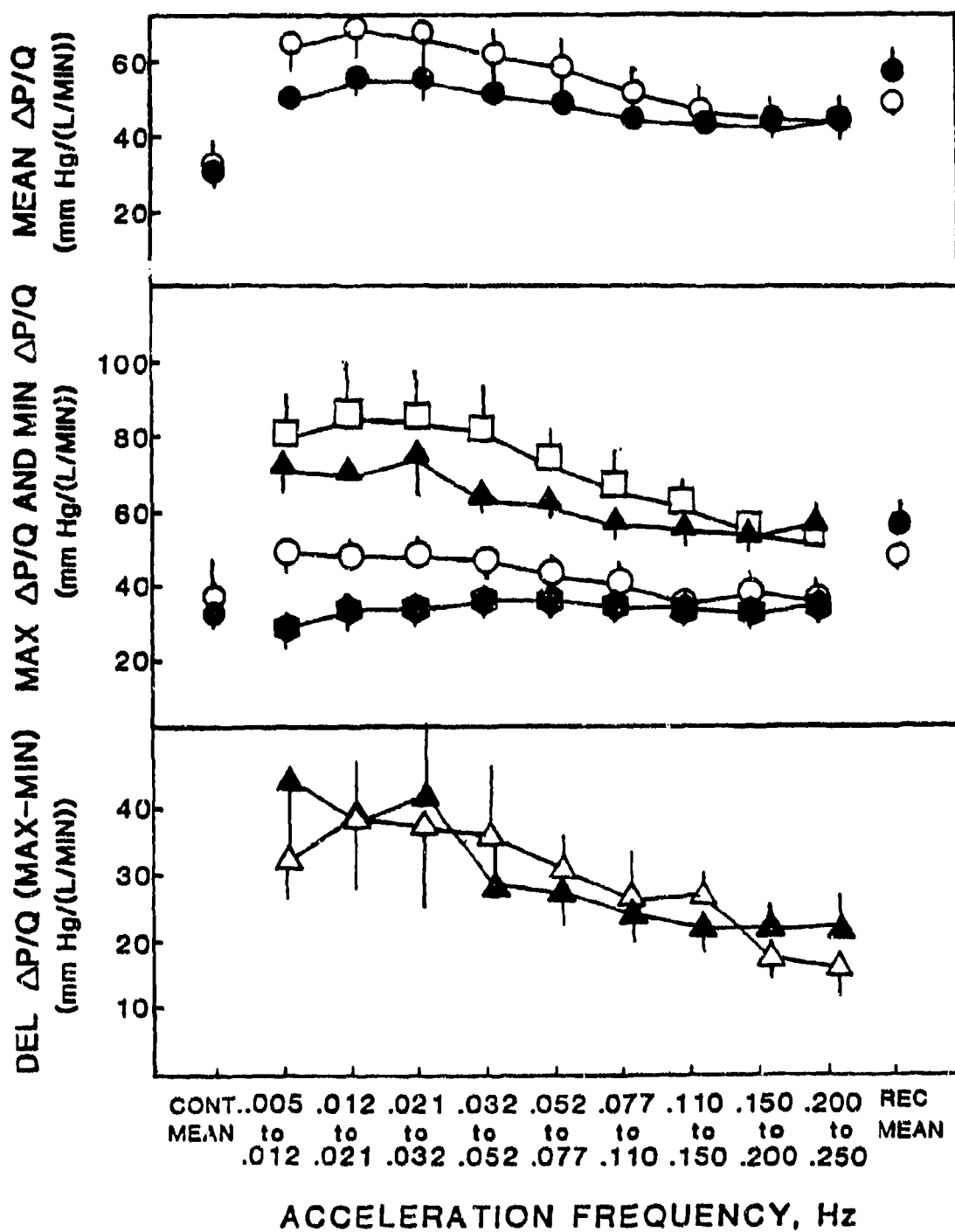


FIGURE II-4. Comparison of the peripheral vascular resistance ( $\Delta P/Q$ ) to acceleration for 5 dogs after autonomic effector blockade (open symbols) and ganglionic blockade (closed symbols).

resistance during the  $+g_z$  portion of the cycle (middle panel). There was a frequency dependence in the mean resistance (top panel) due primarily to the frequency dependence of the maximum (middle) resistance which diminished with increasing frequency.

#### CONCLUSIONS

1. Addition of the ganglionic blocker hexamethonium (10 mg/kg) to the existing autonomic effector blockade resulted in a lower heart rate and a higher stroke volume with no change in cardiac output, peripheral vascular resistance or mean aortic pressure.
2. Low frequency,  $\pm 2g_z$  acceleration produced a post acceleration mean pressor response in both ganglionically blocked and effector blocked dogs. In both groups of dogs, this pressor response was due to an increase in peripheral resistance and was accompanied by a decrease in cardiac output. The increase in resistance was larger in ganglionically blocked dogs; however, the pressor response was not as large since there was a greater decrease in cardiac output. The decrease in cardiac output was due solely to a decrease in stroke volume, with no change in heart rate.
3. Across the frequency range studied, for both groups, peripheral resistance was at its maximum values at the lowest frequencies decreasing with increasing frequency back toward, but never reaching, control values.
4. The response of cardiac output (via stroke volume mechanisms alone) was opposite to that of peripheral resistance: cardiac outputs dropped to the lowest values at the lowest frequencies

and increased with increasing frequency back toward, but never reaching, the pre-acceleration control values.

5. The size of pressure oscillations (DEL AP) decreased with increasing frequency in both groups as a result of decreasing oscillations in both peripheral resistance and stroke volume. This particular trait of the blocked animals appears to describe the actual hydraulic response of the nonreflexive cardiovascular system.

### III. THE EFFECT OF CARDIAC DENERVATION ON CARDIOVASCULAR FUNCTION BEFORE AND AFTER AUTONOMIC EFFECTOR BLOCKADE

#### RATIONALE

After several years of study involving both neurally intact and cardiac denervated dogs, an analysis of the hemodynamic and neurohormonal results of cardiac denervation has been performed. Hemodynamic data was taken during the preacceleration control state at each stage of autonomic blockade (unblocked, following beta blockade, following addition of muscarinic blockade to the beta blockade, and finally following addition of alpha blockade to the beta and muscarinic blockades). Neurohormonal data, with corresponding hemodynamic data, was taken for the two groups in the unblocked state and following the combined blockade.

#### METHODS

##### Surgical:

a. Cardiac denervation. In fourteen of the thirty one dogs used in this study, the method of Randall (11) was used for denervation of the heart prior to implantation of instrumentation detailed below. The efficacy of the denervation was confirmed prior to chest closure by demonstrating the complete absence of change in atrial and ventricular contractile force and heart rate during stimulation of the left and right thoracic vagi and left and right stellate ganglion, all of which could be visualized through the left thoracic incision used in the implant procedure. On the day of the experiment, denervation was confirmed by the absence of reflex cardiac response to right atrial injections of 8 ug/kg of nitroglycerine (Nitrostat) and 40 ug/kg of phenylephrine (neosynephrine).

b. Implant procedures. Both normal and cardiac denervated dogs underwent the same implant procedure. Each was anesthetized with sodium thiopental (20mg/kg) and was prepared for sterile surgery. A thorocotomy was performed



at L-4 and a pressure gauge (Konigsberg Instruments, model P19) was placed through the apex of the heart into the left ventricular chamber. An electromagnetic flow cuff (Zepeda Instruments) was placed on the ascending aorta and a polyvinyl cannula was placed through the right atrial appendage. Three weeks later, on the day of study, manometer-tipped catheters (Millar PC 350, 5 French) were placed, under Innovar tranquilization (1.5 ml/20 kg, IM) and local anesthetic, in the right and left ventricles of the heart via small branches of the femoral vein and artery. The arterial gauge was used to calibrate the implanted gauge and was then retracted into the aorta, just outside the valve, to record arterial pressure for the remainder of the study.

c. Postoperative care. The surgery, recovery and experiments were conducted in accordance with those procedures outlined in the "Guiding Principles in the Care and Use of Animals". Each animal was allowed a minimum of three weeks of postoperative recovery before studies were begun. Postoperative management included antibiotic coverage and particular attention to body temperature and nutritional and hemotologic factors. Studies were not performed unless these factors were stable and within a range considered to indicate a satisfactory state of health.

Experimental Protocol and Blood Sampling Techniques:

Right atrial blood samples were withdrawn from twenty four animals (13 normal and 11 denervated ) about 3 minutes before 30 to 40 minutes of sinusoidal acceleration stress. Samples were withdrawn in both the 'reflexive' state and following total autonomic blockade, detailed below. In addition to hematocrit, each sample was analyzed for at least two of the following substances: plasma renin activity, plasma osmolality and arginine vasopressin activity (ADH), plasma catecholamines and/or plasma volume. Subsequent analysis of these samples was performed by various laboratories chosen for their expertise in the analyses to be performed.

- a. Plasma renin activity. Plasma renin activity was determined by Dr. Theodore Kotchin of the University of Kentucky, Department of Medicine.
- b. Plasma arginine vasopressin activity and osmolality. Plasma arginine vasopressin activity and osmolality was determined by Dr. Gary Robertson of the University of Chicago, Department of Endocrinology.
- c. Plasma norepinephrine and epinephrine activity. Plasma norepinephrine and epinephrine activity was determined by Dr. Michael Ziegler of the University of California, San Diego, Department of Medicine.
- d. Plasma volume and hematocrit. Plasma volume was determined by the RISA<sup>131</sup> technique through the Department of Nuclear Medicine, University of Kentucky and hematocrit was done in our own laboratory.

Total Autonomic Efferent Blockade:

Blockade of autonomic effector activity was established in each animal in the same manner. Beta adrenergic activity was recorded in response to a 10 µg injection of isoproterenol (Isuprel), followed by activation of alpha adrenergic responses to a 40 µg/kg injection of phenylephrine (Neosynephrine). Beta adrenergic blockade was then established with a 1 mg/kg infusion of propranolol (Inderal) the dosage used being that required to block any cardiovascular response to a repeated injection of Isuprel.

Next, muscarinic cholinergic blockade was established with a .1 mg/kg infusion of atropine sulfate. Any change in heart rate with the beta blockade already established was then used as an indicator of diminished cholinergic blockade. Finally, alpha adrenergic blockade was established with an hour-long infusion of 30 mg/kg phenoxybenzamine (Diabenzylamine). At the end of this infusion period, second dosages of the beta and the cholinergic blockers were administered, the absence of response to the alpha and beta agonists was verified, blood samples were withdrawn, and 30 to 40 min. of acceleration was begun.

### Data Acquisition

An on-line digital computer sampled all signals at 2-ms intervals during systole and at 8-ms intervals during diastole. Aortic and left and right ventricular pressures were analyzed for peak systolic, mean diastolic and pulse pressures. The program integrated the aortic flow signal after determining flow zero and eliminating pacing and/or ECG artifacts, producing a correct value of stroke volume (ml/beat). Stroke volume was then multiplied by the preceding heart rate (beats/min) and converted to an analog signal to give a one-beat-delayed, beat-by-beat strip-chart recording of cardiac output (ml/min). Using values stored from an individual beat, the peripheral vascular resistance was calculated with the use of the pressure difference from the aortic valve to the tricuspid valve (mean aortic pressure --diastolic right ventricular pressure), divided by cardiac output (stroke volume X heart rate). Each computed variable was converted to an analog signal, sent to analog tape for later analysis, to the strip-chart recorder for continuous monitoring and to digital tape for summary over each control, test and recovery period.

### Statistical Analysis:

The change in aortic blood pressure at each stage of the autonomic blockade for either the normal or cardiac denervated dogs was tested statistically with a one-way analysis of variance followed, if indicated, by a Duncan Multiple Range test to indicate which means were significantly different. Likewise, changes in heart rate, stroke volume, cardiac output, peripheral resistance, hematocrit, plasma renin activity, plasma levels of vasopressin and plasma levels of catecholamines were tested for significance using the same analysis. Differences between normally innervated and cardiac denervated animals at each stage of the blockade was tested with an analysis of variance for unequal number of subjects. Post hoc testing in this case was done with

the t-test for differences among several means.

## RESULTS

### a. Unblocked vs Total Autonomic Effector Blockade

Comparison of hemodynamic variables from 8 normally innervated and 8 cardiac denervated dogs are shown in figure III-1. The first pair of bars for each variable are for the unblocked animals and the second pair are for the same animals following blockade of  $\alpha$  and  $\beta$  adrenergic and muscarinic receptors. Triangles indicate significant changes in either group as a result of blockade of autonomic effector activity, while circles indicate significant differences between the normally innervated and cardiac denervated groups for a given state (unblocked or autonomically blocked). Aortic pressure for the two groups in the unblocked state is essentially the same and is lowered significantly to the same level in both groups by autonomic blockade. Similarly, peripheral vascular resistance is lowered significantly in both groups by the blockade, but there was no difference between groups. Mean left ventricular volume was slightly but not significantly elevated as a result of cardiac denervation and was further elevated in both groups (again not significantly) by autonomic blockade. Cardiac output was identical in both groups in the unblocked state, did not change in the normally innervated animals as a result of blockade, and was slightly but not significantly lowered in the cardiac denervated animals with blockade. Stroke volume, like aortic pressure and peripheral resistance, was the same in both groups in the unblocked state and was lowered by blockade in both groups but there was no difference between the two groups.

Heart rate was the only hemodynamic variable measured in which a difference between normally innervated and cardiac denervated dogs appeared, and that difference was not evident until autonomic blockade was established.

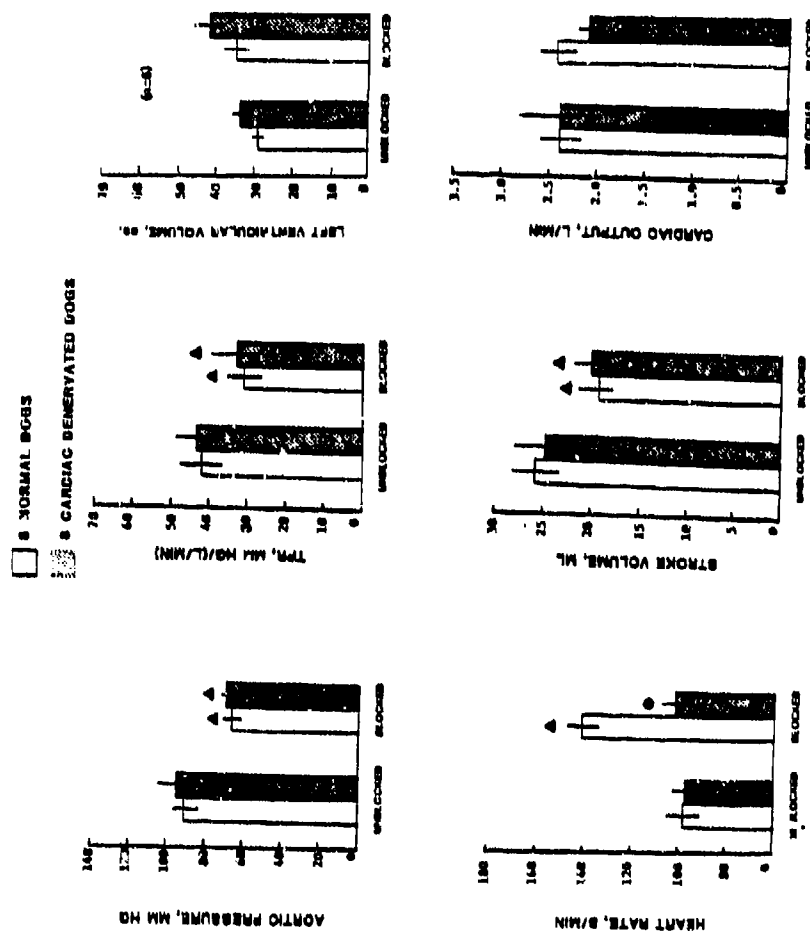


FIGURE III-1. Mean  $\pm$  S.E.M. value of hemodynamic variables for 8 normally innervated and 8 cardiac denervated dogs in the unblocked state and following blockade of  $\alpha$ ,  $\beta$  and muscarinic effector activity. Triangles indicate significant changes in a variable as a function of blockade. Circles indicate significant differences between normal and denervated dogs for that variable.

In the unblocked state, heart rates were  $98 \pm 6$  b/min for both groups; in normally innervated dogs, heart rates rose significantly to  $140 \pm 6$  b/min following blockade of  $\beta$  and  $\alpha$  adrenergic and muscarinic receptors, while in denervated dogs, heart rates rose to  $105 \pm 6$  b/min following blockade.

The neuro-hormonal variables measured under the same conditions are shown in Figure III-2. Again, triangles indicate significant changes in a variable as a result of autonomic blockade and circles indicate a significant difference between normally innervated and cardiac denervated dogs in a given state.

Hematocrit was significantly elevated in cardiac denervated dogs and was not changed in either group by autonomic blockade. To go with this, but not shown, was a decrease (nonsignificant) in plasma volume in 4 cardiac denervated dogs (820 ml) as compared to 5 normally innervated dogs (1100 ml) in the unblocked state with no apparent change in either group as a result of autonomic blockade. Plasma renin activity, already low (2.1 ng/ml/hr) in unblocked, normally innervated dogs was decreased by cardiac denervation to  $<1$  ng/ml/hr. Blockade of autonomic effector activity significantly elevated plasma renin activity in both groups but the difference between the two groups was no longer significant. Plasma arginine vasopressin activity was not significantly different between the two groups in either the unblocked or autonomically blocked states, however the blockade significantly elevated vasopressin activity in both groups. Plasma osmolality, like vasopressin, was the same for both groups in both states, but the blockade significantly elevated osmolality in both groups. Plasma norepinephrine was significantly lowered by cardiac denervation in the unblocked state and was significantly elevated in both groups by the blockade. There were no significant plasma epinephrine differences between the groups or with autonomic blockade, however

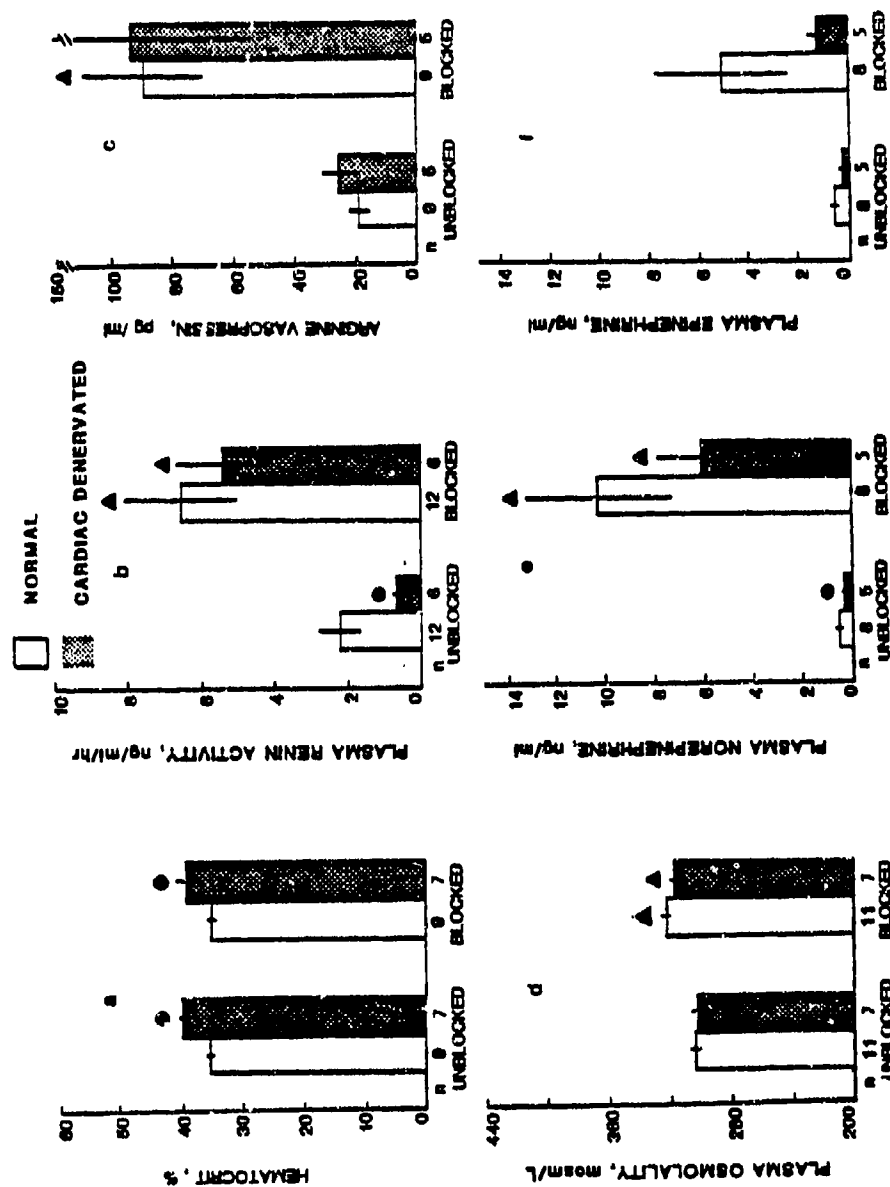


FIGURE III-2. Mean  $\pm$  S.E.M. values of neuro-hormonal variables for a group of normally innervated and a group of cardiac denervated dogs in the unblocked state and following blockade of  $\alpha$ ,  $\beta$  and muscarinic effector activity. Triangles indicate significant changes in a variable as a function of blockade. Circles indicate significant differences between normal and denervated dogs for that variable.

taking the logarithm of the values before testing for significance indicated a lower value of epinephrine in cardiac denervated dogs and an increased value in normally innervated dogs as a result of the blockade.

b. Dissection of the hemodynamic response to each step of the autonomic blockade

The results of the comparison of unblocked animals to animals following autonomic effector blockade of  $\beta$  and  $\alpha$  adrenergic and muscarinic effector activity led to an analysis of the response after each stage of the blockade. The results of this analysis are shown in Figures III-3 and III-4 for normally innervated dogs (bars 1-4) and cardiac denervated animals (bars 5-8). Data are shown for the unblocked state (bars 1 & 5 lightest stippling), after blockade of  $\beta$  adrenergic activity with 1mg/kg propranolol (bars 2 & 6), after addition of muscarinic blockade with 0.1 mg/kg atropine to the  $\beta$  blocked animal (bars 3 & 7), and finally following addition of  $\alpha$  blockade with 20-30 mg/kg phenoxybenzamine to the  $\beta$  + muscarinic blocks (bars 4 & 8). Triangles indicate a significant change from the preceding state, unless otherwise indicated, as in  $\max d(LVP)/dt$ , and stars indicate a significant difference between normally innervated and cardiac denervated animals in the same state.

Aortic pressure was not different for 13 normally innervated and 13 cardiac denervated dogs nor did pressure change significantly from the unblocked state in either group until the administration of the  $\alpha$  adrenergic blocker. Total peripheral resistance was not significantly different between the two groups at any stage of blockade, however administration of the  $\beta$  adrenergic blocker did elevate peripheral resistance in both groups and addition of the  $\alpha$  blocker significantly lowered resistance in both groups. There was no difference in cardiac output between normally innervated and cardiac denervated animals at any stage of blockade, however  $\beta$  blockade significantly



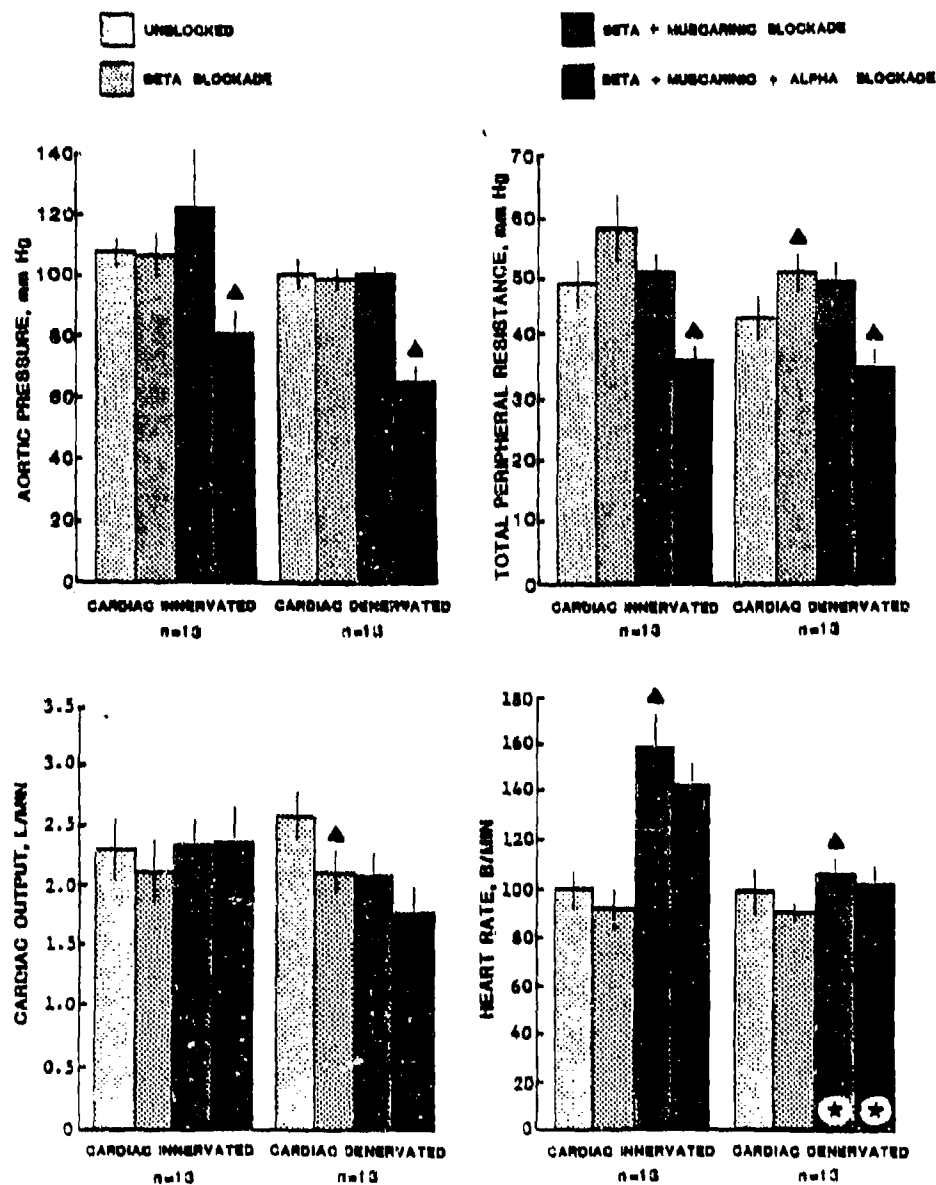


FIGURE III-3. Mean  $\pm$  S.E.M. values of hemodynamic variables for a group of 13 normally innervated and 13 cardiac denervated dogs at each stage of blockade of autonomic effector activity. Triangles indicate a significant change from the preceding blocked state. Stars indicate a significant difference between normal and cardiac denervated groups for that particular state of blockade.

lowered cardiac output in denervated dogs. Subsequent blockade of  $\alpha$  receptors further lowered (N.S.) cardiac output in denervated dogs while slightly elevating (N.S.) it in normally innervated dogs. The components of cardiac output, heart rate and stroke volume are the variables in which a difference was evident between normally innervated and cardiac denervated hearts. In the unblocked state, heart rates were the same for both groups and there was not significant change in either group with the addition of the  $\beta$  blocker. Addition of the muscarinic blockade increased heart rates significantly in both groups but in normally innervated dogs heart rates rose to ~ 160 b/min while denervated dogs only rose to 108 b/min and the difference between the groups was significant. Addition of  $\alpha$  blockade did not change the significant difference between the normally innervated and denervated hearts. Stroke volumes were lowered significantly by  $\beta$  blockade in denervated hearts and by the condition of  $\beta$  and muscarinic blockade in normally innervated hearts. In the  $\beta$  + muscarinic state, cardiac denervated dogs maintained a significantly higher stroke volume than did the normal dogs.

The determinants of stroke volume, mean left ventricular volume (an index of left ventricular diastolic volume),  $\max d(LVP)/dt$ , and right ventricular diastolic pressure are shown in the remaining three sections of III-4. Mean left ventricular volume for 8 normally innervated and 6 cardiac denervated dogs increased nonsignificantly with succeeding blockades in cardiac denervated dogs and increased greatest in normally innervated dogs with the  $\beta$  blockade. Maximum  $d(LVP)/dt$  decreased significantly in both groups with  $\beta$  blockade and decreased significantly further with the addition of muscarinic and  $\alpha$  blockades in 8 denervated dogs and with the addition of  $\alpha$  blockade to  $\beta$  + muscarinic blockades in 8 normally innervated dogs. Right ventricular diastolic pressure increased significantly in both groups with  $\beta$  blockade,

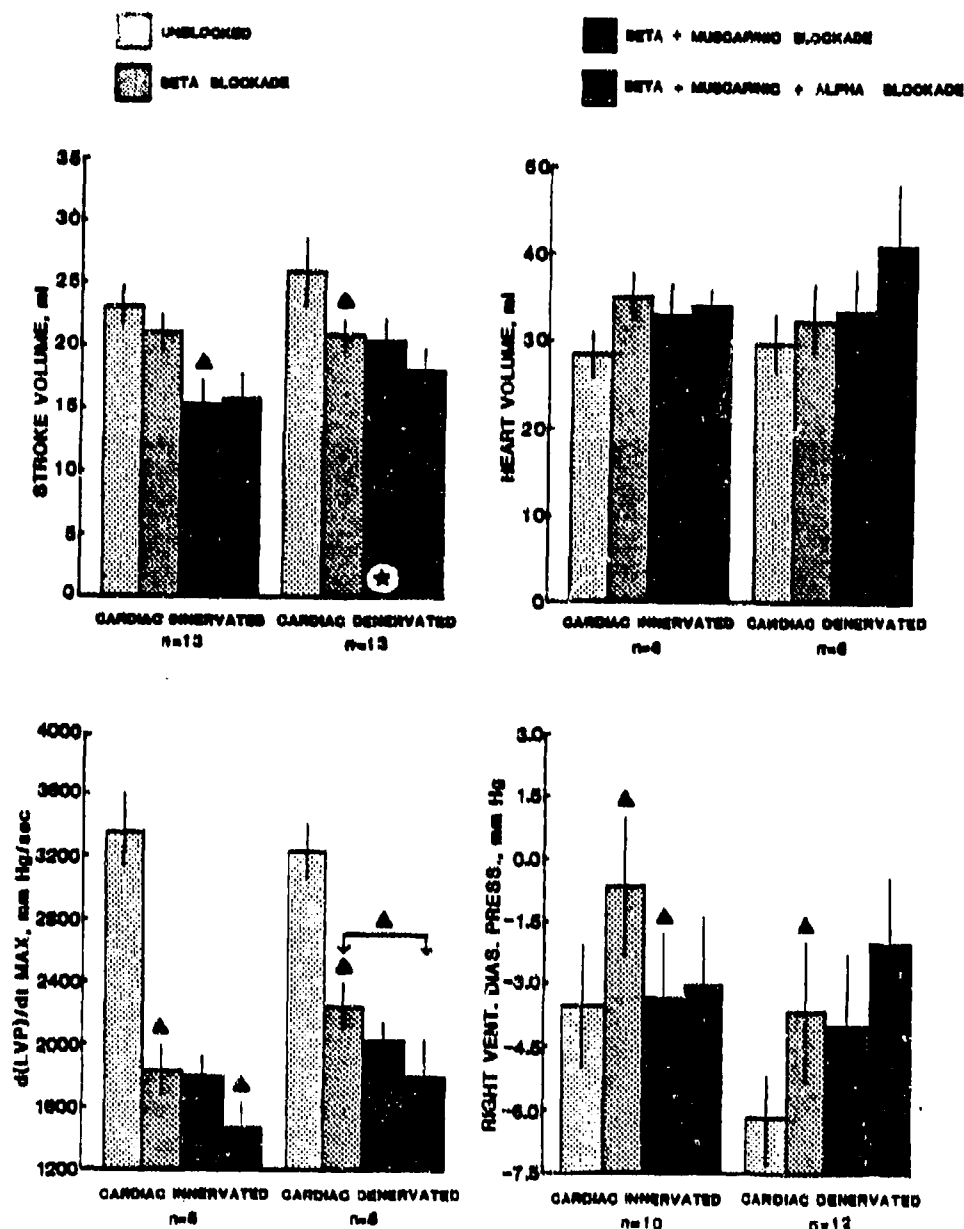


FIGURE III-4. Mean + S.E.M. values of hemodynamic variables for a group of normally innervated and a group of cardiac denervated dogs at each stage of blockade of autonomic effector activity. Triangles indicate a significant change from the preceding blocked state. Stars indicate a significant difference between normal and cardiac denervated groups for that particular state of blockade.

and decreased significantly in the normally innervated dogs with the addition of muscarinic blockade.

#### DISCUSSION AND CONCLUSIONS

The effect of three weeks of cardiac denervation on hemodynamic and neurohormonal variables was examined in the unblocked, but tranquilized, state and in the same animals following blockade of autonomic effector activity. The data from this group of animals were compared to those from a control group of neurally intact animals under the same conditions. The principle conclusion to be drawn from this study is that cardiac denervation has minor but not drastic effects on hemodynamic or neurohormonal variables in the control state and for that reason a comparison of the responses of the two groups to sinusoidal acceleration stress (Sect. I) is reasonable.

In the first part of the study (Figures III-1 and III-2), it was determined that for unblocked animals aortic pressure, cardiac output, stroke volume, peripheral resistance, heart rate, plasma vasopressin, osmolality and epinephrine showed no statistical difference in the normally innervated as compared to the cardiac denervated groups of animals. However, cardiac denervation significantly increased hematocrit and significantly decreased plasma renin activity and norepinephrine in these unblocked animals.

Following blockade of autonomic effector activity, heart rates were significantly elevated in normally innervated dogs as compared both to this groups' unblocked values and to blocked values in cardiac denervated dogs. This was the only hemodynamic difference seen between the two groups, however there were significant changes in both groups as a result of blockade; aortic pressure, peripheral resistance and stroke volume were lowered. Similarly there were dramatic changes in neurohormonal variables as a result of the blockade, but no further differences between the two groups developed.

Hematocrit did not change in either group with blockade while plasma renin, vasopressin, osmolality, norepinephrine and (N.S.) epinephrine rose in both groups as a result of the blockade.

In the next part of the study [Figures III-3 and III-4] a comparison of changes at each stage of blockade and between normal and denervated dogs was made for an expanded group of hemodynamic variables. Beta blockade was found to significantly lower stroke volume,  $d(LVP)/dt$  max, and cardiac output and raise heart volume (NS) peripheral resistance and diastolic right ventricular pressure in both groups. Addition of muscarinic blockade significantly elevated heart rate in both groups but the rate in neurally intact animals went to 160 b/min while in cardiac denervated animals it only went to 108 b/min. This increase in rate in both groups is probably responsible for the decrease in right ventricular diastolic pressure that was significant in the normal dogs but not in the denervated dogs. Also at this level of blockade the first statistically significant differences between the two groups appear; the increased heart rate in neurally intact dogs which was offset by a decreased stroke volume in the same dogs leaving cardiac output unchanged. Addition of the alpha blocker significantly decreased aortic pressure, total peripheral resistance and unexpectedly, max  $dp/dt$ . The decrease in max  $dp/dt$  combined with the increase in heart volumes (N.S.) and the low stroke volume tends to indicate that the ventricles of both groups are working at a greater than optimal end diastolic fiber length.

The conclusions from this phase of the study are listed below.

#### Cardiac Denervation Conclusions

1. Unblocked "reflexive" dogs
    - a. Lower plasma renin activity
    - b. Lower plasma norepinephrine activity
    - c. Lower plasma epinephrine activity
    - d. Lower plasma volume
- } significant  
 } approach significance

2. Autonomically blocked "nonreflexive" dogs
  - a. Lower "intrinsic" heart rate that was significant only after combined beta and muscarinic blockade.
  - b. Higher stroke volume in the beta and muscarinic blocked state possibly due to the lower heart rates in these animals (due to the inverse relationship between heart rate and stroke volume at high heart rates).
  - c. Lower plasma volume, renin, norepinephrine, epinephrine that were no longer significant after the blockade was established.

#### Autonomic Blockade Conclusions

1. Aortic pressure was significantly lowered by alpha blockade due to the lowering of peripheral resistance.
2. Peripheral resistance was elevated significantly by beta blockade in cardiac denervated dogs and non significantly in normal dogs.
3. Cardiac output was significantly lowered by beta blockade in cardiac denervated animals.
4. Heart rate was significantly elevated by combined beta and muscarinic blockades in both groups of animals.
5. Heart volume was elevated, but not significantly, in both groups by combined blockade indicating that the diminished stroke volume in both groups after blockade was due to loss of effector activity rather than diminished end diastolic fiber length.
6. Significantly diminished stroke volume was apparent in cardiac denervated dogs after beta blockade and in normal dogs after combined beta and muscarinic blockades.
7. Maximum  $d(LVP)/dt$  was significantly lowered by beta blockade in both groups and was further lowered by the subsequent blockades.
8. Right ventricular pressure was significantly elevated by beta blockade in both groups and was significantly lowered by the addition of muscarinic blockade in normal dogs, perhaps due to the increased heart rate accompanying this blockade.
9. Hematocrit and plasma volume were unaffected by blockades.
10. Plasma renin activity, norepinephrine, vasopressin, osmolality and epinephrine (marginally) were significantly elevated by the addition of the alpha blockade to the beta and muscarinic blockade. The sequential changes with each blockade were not shown but no increase in any of these variables occurred until the alpha blocker was added.

## APPENDIX A

RESEARCH TEAMInvestigators

C. F. Knapp, Ph.D.	Department of Mechanical Engineering, Wenner-Gren Biomedical Engineering Laboratory
D. Randall, Ph.D.	Department of Physiology and Biophysics
J. Evans, M.S.	Wenner-Gren Biomedical Engineering Laboratory
B. Kelley, Ph.D. candidate	Department of Mechanical Engineering
J. Charles, Ph.D. candidate	Department of Physiology and Biophysics
D. Bhavsar, M.S. candidate	Department of Mechanical Engineering

Collaborators

W. Griffen, M.D.	Department of Surgery, University of Kentucky Medical Center
T. Kotchen, M.D.	Department of Medicine, University of Kentucky Medical Center
G. Robertson, Ph.D.	Department of Endocrinology, University of Chicago
M. Ziegler, Ph.D.	Department of Medicine, University of California, San Diego

Technical Staff

Surgical Technicians:	C. Woolfolk, B. Rockwood, D. Graham and L. Ennis
Instrumentation Specialists:	R. Stanifer and A. Liaupsin
System and Data Analysts:	E. Lowery, C. Fischer, G. Hirsch, B. Seelye, E. Cremers, K. Kelley, J. Funk

The formation of this research team is based on a general plan which integrates the advanced analytical techniques and instrumentation development capabilities of an interdisciplinary team, consisting of physiologists and

biomedical engineers, in an effort to resolve problems associated with acceleration stress. Measurements from the invasive instrumentation of the chronically implanted animal preparation of this study are essential for identifying the most meaningful variables for assessing acceleration-induced cardiovascular responses when less invasive measurements are eventually to be made on man. It is our belief that this basic research effort will provide the background for the design and implementation of human investigations, investigations which will lead to improved protective equipment and operational procedures for military personnel exposed to acceleration environments resulting from the optimal utilization of advanced aerospace systems.



## PRESENTATIONS

Knapp, C. F., J. M. Evans, D. C. Randall. Cardiac dimension changes in canines during low frequency sinusoidal acceleration ( $< 0.25$  Hz). Meeting on "Research Methods in Human Motion and Vibration Studies. New Orleans, September 1981.

Evans, J. M., D. C. Randall and C. F. Knapp. The effect of cardiac denervation on cardiovascular function before and after autonomic blockade. ABSTRACT #612 APS Fall Meeting, October 1981.

Vallance, S. R., T. S. Skinner, G. E. Billman, C. L. Fischer, D. C. Randall, C. F. Knapp and J. M. Evans. Effects of fentanyl-droperidol (Innovar) on hemodynamic responses to acute coronary occlusion in intact dog. ABSTRACT #100 APS Fall Meeting, October 1981.

Charles, J. B., D. R. Richardson, D. C. Randall, C. F. Knapp and J. M. Evans. Changes in cardiovascular responses to low-frequency sinusoidal accelerations with endurance training and detraining in dogs. Proceedings IEEE 1981 National Aerospace and Electronics Conference. Vol 2, 581-583, Dayton, OH, May 1981.

## PUBLICATIONS

Knapp, C. F., J. M. Evans, D. C. Randall and J. A. Marquis. Regulation of blood pressure and cardiac function in canines during low frequency acceleration. Accepted by the Am. J. Physiol. (Heart and Circ.), (Submitted August 1981).

Evans, J. M., C. F. Knapp and D. C. Randall. Neurohormonal interactions in mediation of an acceleration induced pressor response. Submitted to Am. J. Physiol. (Heart and Circ.), September 1981.

Randall, D. C., J. M. Evans, G. E. Billman, G. A. Ordway and C. F. Knapp. "Neural Hormonal and Intrinsic Mechanisms of Cardiac Control During Acute Coronary Occlusion in Intact Dog". J. Autonom. Nerv. Sys., 3(181) 87-99.